NATIONAL INSTITUTE OF SIDDHA

TAMBARAM SANATORIUM, CHENNAI - 47

THE TAMIL NADU Dr. M.G.R. MEDICAL UNIVERSITY

CHENNAI – 32

Pre-clinical and clinical study on Valendraphola Chooranam for Haematinic Activity in the management of Pandu (Anaemia)

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Pre-clinical and clinical study on Singathi Chooranam for Bronchodilator Activity in the management of Eraippu (Bronchial Asthma)

(DISSERTATION SUBJECT)

For the partial fulfillment of the

requirement to the Degree of

DOCTOR OF MEDICINE (SIDDHA)

BRANCH II - GUNAPADAM

APRIL – 2013

TRIAL DRUG I: VALENDRAPHOLA CHOORANAM

TRIAL DRUG II: SINGATHI CHOORANAM

INTRODUCTION

Anaemia is one of the common blood disorders. It refers to the reduction in red blood cell count, haemoglobin contents, packed cell volume. Basically it is a condition when the haemoglobin content of blood decrease below normal level[1].

Generally, Anaemia occurs because of decrease in the production of RBC, increased destruction of RBC and excess loss of blood from the body. Iron deficiency anaemia is the most common type of anaemia. It develops due to inadequate availability of iron for haemoglobin synthesis. Causes of Iron deficiency anaemia are loss of blood, decreased intake of iron, poor absorption of iron from intestine, increase in demand for iron in conditions like growth and pregnancy. Iron deficiency anaemia is the most common form of nutritional deficiency affecting adults in rural as well as urban area. In different age group who are subjected to numerous stress which affects their health and well being[1].

The WHO estimates that more than 1/3rd of the world population is Anaemic, of which Iron deficiency anaemia is the most common and serious problem of public health significance. In India Prevalence of Anaemia is the highest in the world but within the country prevalence rates differ substantially between different regions[2].

Globally, Anaemia affects 1.62 million people which corresponds to 24.8% of the Population. The highest prevalence is in preschool-age children (47.4%) and the lowest prevalence in men (12.7%). However, the population group with the greatest number of individuals affected is non-pregnant women (468.4 million/30.2%)[3].

National Family Health Survey has reported anaemia prevalence of 56.2 percent in women of 15-49 yr and 24.3 percent in men aged 15-49 yr. Iron deficiency is the most common cause of anaemia in the world affecting 30% of the world's population equivalent to 500 million people[4].

Prevalence of anaemia in rural populations in Tamil nadu is as high as 52%. This could be explained due to lack of awareness in these individuals[5]. 60-70% of Indian adolescent girls are anaemic. In children and young adults particularly in deprived socio-economic group, the prevalence of iron deficiency anaemia is 5% and 10% respectively[6].

In Siddha system of medicine, Anaemia is compared to Pandu noi. According to our text, Pandu noi has 5 types. In which, the symptoms of Pitha Pandu coincides with the symptoms of Iron deficiency anaemia. (Paleness of the tongue, fatigue, dyspnoea, giddiness, oral ulcer)[7]. According to the Siddha Medicine, when the normal equilibrium of the three humors (Vatham, Pitham, Kabam) is deranged, disease occurs. In Pandu noi, the Pitham and Kapam is deranged.

The preparation "Valendraphola Chooranam" mentioned in the Siddha text, “Gunapadam mooligai vaguppu" is indicated for Pandu. The drug Valendrapholam is bitter in taste[8]. The bitter taste has the property of neutralising pitham and kapam, thereby it helps in treating Pandu.

The previous studies of Valendrapholam shows the property of antioxidant, immuno-modulator, astringent, sedative.

Hence the author have selected Valendraphola Chooranam for Pandu.

AIM:

To evaluate the safety and efficacy of Valendraphola Chooranam for Haematinic activity in the management of Pandu (Anaemia).

OBJECTIVE:

Primary objective:

To evaluate the Haematinic activity of "Valendraphola Chooranam" for "Pandu" (Anaemia) in preclinical studies.

Secondary objective:

Biochemical analysis.

To evaluate the efficacy of Valendraphola Chooranam [Commiphora myrrha] in the management of Pandu [Anaemia].

High performance thin layer chromatography.

MATERIALS AND METHODS

STANDARD OPERATIVE PROCEDURE:

COLLECTION AND AUTHENTICATION OF RAW DRUG:

The raw drug was procured from Ramasamy chetty shop, Paris, Chennai, and authenticated by competent authority in Department of Gunapadam, National Institute of Siddha, Chennai.

INGREDIENTS:

Valendrapholam (Commiphora myrrha)

PURIFICATION PROCESS[9]:

Purification of Valendrapholam:

The raw drug was purified by boiled in the vinegar.

PREPARATION OF THE MEDICINE[8]:

The raw drug was purified and then dried and pulverized by an electric grinder into fine powder. And then it was sieved by using a fine silk cloth (vasthrakayam). The powder was stored in a clean, dry, air tight glass container.

LABELLING :

Name of the Preparation	: Valendraphola Chooranam
Quantity of the drug	: 14 gm
Dose	: 1 gm bid
Adjuvant/Vehicle	: Luke warm water
Indication	: Pandu
Date of manufacturing	: The drug was prepared in two batches. 23/05/2012, 20/08/2012
Date of expiry	: 3 months from the date of manufacture.

வாலேந்திரபோளம்

Commiphora myrrha

(BEFORE PURIFICATION) **(AFTER PURIFICATION)**

VALENDRAPHOLA CHOORANAM

REVIEW OF LITERATURE

GUNAPADAM ASPECT

வாலேந்திரபோளம்[8]

வேறுபெயர் : குங்குமதீபம், குந்துரு, மீறு, வெள்ளைப்போளம், வெள்ளாத்திபோளம்

பயன்படும் உறுப்பு : பிசின்

சுவை : கைப்பு

தன்மை : வெப்பம்

பிரிவு : கார்ப்பு.

செய்கை :

வெப்பமுண்டாக்கி
கோழையகற்றி
ருதுவுண்டாக்கி
பசித்தீத்தூண்டி
அகட்டு வாய்வகற்றி

குணம் :

“சூலைகய ரோகம் சொறிகரப்பான் குன்ம மிவை
ஆலைவா யின்கோல்போல் ஆகுங்காண் - நீலசக்கர
வாளமெனத் தோற்றுமுலை மாதரசே! - வாலேந்திர
போளந் தனையெடுக்கும் போது.”

இதனால் கீல்பிடிப்பு, இளைப்பு நோய், கரப்பான், குன்மம், சூதகக்கட்டு, சூதகப்பாண்டு, பாண்டு, இரைப்பு, மந்தம், வாய்ப்புண் முதலியவை தீரும்.

வாலேந்திரபோளம் சேரும் மருந்துகள் :

பிரசவ ரஷாமிர்த மாத்திரை10:

அளவு : 1 மாத்திரை, 2 வேளை

தீரும் வியாதி : பிரசவித்த ஸ்திரீகளுக்கு, உதிரக்கட்டை தடையின்றி வெளிப்படுத்தும், எவ்விதப் பிணியையும் வர வொட்டாது.

மூசாம்பிர மெழுகு[10]:

அளவு : 2 கடலை பிரமாணம், 5 நாள்.

தீரும் வியாதி : ஸ்திரீகளுக்கு மாத விலக்குக் காலத்தில் உண்டாகும் வயிற்றுவலி குணமாகும். சந்தான சித்தியுண்டாகும்.

BOTANICAL ASPECT

VALENDRAPHOLAM[11]

BOTANICAL NAME: Commiphora myrrha

VERNACULAR NAMES[12]:

ENG	: Myrrh
SANS	: Gandha-rasaha
HINDI	: Bola
TEL	: Balintra-polamu
KAN	: Bola

SCIENTIFIC CLASSIFICATION:

Kingdom	: Plantae
Order	: Sapindales
Family	: Burseraceae
Genus	: Commiphora
Species	: myrrha

Botanical name: Commiphora myrrha

HABITAT:

"Indigenous to North-Eastern Africa." It is collected in Southern Arabia, Abyssinia, Persia, Siam and sold in Indian Bazaars. Myrrh of commerce is obtained from the resinous exudation of the tree Commiphora myrrha. There are at least two or three varieties, two of them being known as 'Karam' and 'Mutiya'.

PARTS USED:

Gum from the bark of the tree.

CHEMICAL CONSTITUENTS:

Myrrhol

Myrrhic acid

Calcium phosphate, Carbonate

Cumic aldehyde

Eugenol

Meta-cresol

Pinene

di-pentene

Limonene

ACTIONS:

Stimulant

Expectorant

Emmenogogue

Astringent

USES:

It is useful in dyspepsia and mixed with molasses or preferably with iron and vegetable bitters it is given in amenorrhoea, chlorosis.

It is a good stomachic and laxative.

PHYSICAL PROPERTIES

Materials and Methods

The Physical properties of Valendraphola Chooranam were analysed in the following procedure. It was done at Sri Ramachandra University, Chennai.

pH at 10% of aqueous solution:

Five grams of Valendraphola Chooranam was weighed accurately and placed in clear 100 ml beaker. Then 50 ml of distilled water was added to it and dissolved well. After 30 minutes it was then applied in to pH meter at standard buffer solution of 4.0,7.0,9.2. (Trial drug I,Table 2)

Ash Values

The Ash values measures inorganic constituents present in the raw drug. A high ash content explains its unsuitable nature to be used as a drug. (Trial drug I, Table 2)

Total Ash

A little of extract was taken in a silica crucible previously ignited, cooled and weighed. It was incinerated by gradually increasing the heat not exceeding dull red heat (450°C) until free from carbon, cooled and weighed. The percentage of ash was calculated with reference to air-dried drug. The procedure was repeated to get the constant weight. (Trial drug I, Table 2)

Water soluble ash

The total ash was boiled with 25 ml water and filtered through ash less filter paper (Whatmann 4.1). It was followed by washing with hot water .The filter paper was dried and ignited in the silica crucible, cooled and the water insoluble ash was weighed. The water-soluble ash can be calculated by subtracting the water insoluble ash from the total ash. (Trial drug I, Table 2)

Acid insoluble ash

The total ash obtained was boiled for 5 minutes with 25 ml of (10% w/v) dilute hydrochloric acid and filtering through ash less filter paper (Whatmann 4.1). the filter paper was ignited in the silica crucible, cooled and insoluble ash was weighed. (Trial drug I, Table 2).

HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY

The HPTLC of Valendraphola Chooranam was done at Sri Ramachandra University, Chennai.

HPTLC Fingerprint - RH1

SAMPLE PREPARATION

100 mg of extract was weighed and dissolved in 70% methanol to get a concentration of 10 mg/ml concentration this is then used for injection.

CHROMATOGRAPHIC CONDITION FOR HPTLC FINGER PRINT (Trial drug I, Graph 1)

SampleName	:	Valendraphola Chooranam
Sample-ID	:	111
Stationary phase	:	Silica gel F 254
Mobile phase	:	n-Hexane: Ethyl acetate: Formic acid 60:40:2.5 ml)
Scanning wavelength	:	254,298,489 nm
Sample concentration	:	20 mg/ml
Injecting volume	:	5, 10 µl
Development mode	:	Ascending mode

Significance of HPTLC fingerprinting in Standardisation

Standardisation of traditional medicine has become mandatory in the present national and international scientific scenario, as they have to stand competing with stringent regulatory methods and also clinically. HPTLC is one of the versatile chromatographic methods presently available for the rapid analysis of herbal drugs due to several reasons. Firstly the time required for the demonstration of the most of the characteristic constituents of a drug is very quick and short. Secondly, in addition to qualitative detection, HPTLC also provides semi-quantitative information on the major active constituents of a drug, thus enabling an assessment of drug quality. Thirdly the fingerprint obtained is suitable for monitoring the identity and purity of drugs and for detecting adulteration and substitution. Hence in order to check the identity, purity and

standardise the quantity of active principles in the herbal extracts a HPTLC fingerprint of the ingredient medicinal plant used in the formulation has been obtained.

The distribution of phyto-constituents in a plant depends on various factors such as soil, time of collection period of storage, etc. So, it is necessary to standardize the extract being used for pharmacological studies. HPTLC serves as a convenient tool for finding out the distribution pattern of phyto constituents which is unique to each plant. The HPTLC finger-printing profile establishes the identity and purity of the raw drug being used. It helps in the authentification of the plant material.

Chromatographic Conditions

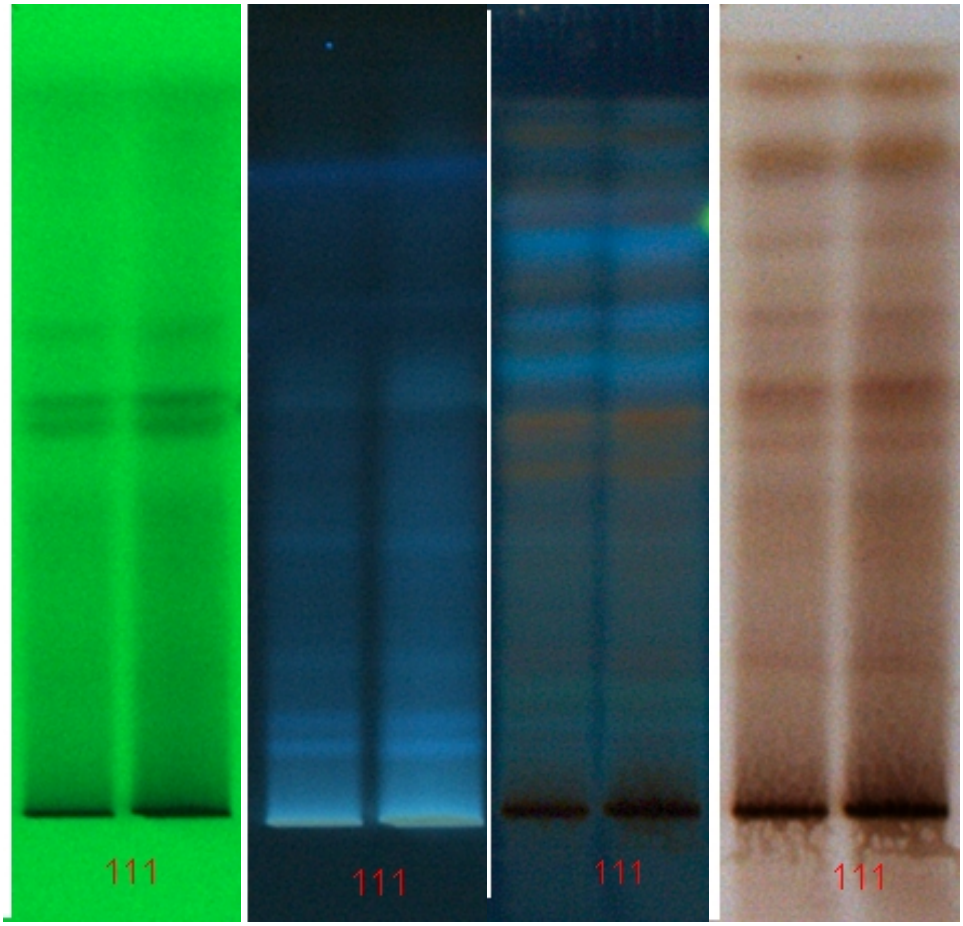
The finger printing has been done using the following chromatographic conditions. Chromatography was performed on a10x10 cm pre activated HPTLC silica gel 60F 254 plate. Samples were applied to the plate as 6mm wide band with an automatic TLC applicator Linomat 5 with N2 flow (CAMAG, Switzerland), 8mm from the bottom. Densitometric scanning was performed on CAMAG scanner III. The plates were pre-washed by methanol and activated at 60^0 C for 5 minutes prior to chromatography. The slit dimension was kept at 5 minutes x 0.45 minutes and 20 minutes scanning speed was employed. The mobile phase was chosen after running each plant in different mobile phases of varying polarity (Toluene, Toluene: Ethyl acetate and Ethyl acetate: Methanol) and 10 ml of mobile phase was used per chromatography. Linear ascending development was carried out in 20 cm x 10-em twin glass chamber saturated with the mobile phase.

Chromatographic Analysis

The hydro alcoholic extracts of the plants have been prepared at a concentration of 10 mg/10 ml in alcohol and were spotted using CAMAG Linomat 5 applicator. The method was optimized by selecting appropriate mobile phase for respective plant extracts and developed in a twin trough chamber, 20 x 10 cm at 25°C. The plates were dried by hair dryer. The developed plates were scanned at appropriate wavelength using CAMAG TLC scanner 3 and photo-documented using CAMAG REPROSTAR 3.

Inference

HPTLC fingerprint of RH -1 shows four peaks at Rf values 0.25, 0.31, 0.41 & 0.95. The peak correspond to the Rf value 0.31 has maximum peak area of 7256.5. At this stage it is difficult to confirm the individual components present in the extract, but from our lab experience on phytochemical analysis, we suggest that the major peaks found in the fingerprint may be acidic glycosides / resins. Since, in the present chromatographic conditions, the above mentioned components will be eluted easy.



254 nm 366nm 366 nm white light

BIO -CHEMICAL ANALYSIS OF VALENDRAPHOLA CHOORANAM

The biochemical analysis of the Valendraphola Chooranam was carried out in the Biochemistry lab, National Institute of Siddha, Chennai.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
1.	Appearance of sample	Light yellow in colour	
2.	**Solubility:** a. A little(500mg) of the sample was shaken well with distilled water. b. A little(500mg) of the sample was shaken well with con. HCl/Con. H_2So_4	Sparingly soluble	Absence of Silicate
3.	**Action of Heat:** A small amount(500mg) of the sample was taken in a dry test tube and heated gartly at first and then strong.	No white fumes evolved	Absence of Carbonate
4.	**Flame Test:** A small amount(500mg) of the sample was made into a paste with con. HCl in a watch glass and introduced into non-luminous part of the Bunsen flame.	No Bluish green flame appeared.	Absence of Copper
5.	**Ash Test:** A filter paper was soaked into a mixture of sample and dil. cobalt nitrate solution and introduced into the Bunsen flame and ignited.	Yellow colour flame appeared.	Absence of sodium

Preparation of Extract:

5gm of Valendraphola Chooranam was weighed accurately and placed in a 250ml clean beaker and added with 50ml of distilled water. Then it was boiled well for about 10 minutes. Then it was cooled and filtered in a 100ml volumetric flask and made up to 100ml with distilled water.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
	I.Test For Acid Radicals		
1.	**Test For Sulphate:** a. 2ml of the above prepared extract was taken in a test tube to this added 2ml of 4% dil ammonium oxalate solution b. 2ml of the above prepared extracts was added with 2ml of dil-HCl was added until the effervescence ceases off. Then 2ml of dil.Barium chloride solution was added.	Cloudy appearance present	Absence of Sulphate
2.	**Test For Chloride:** 2ml of the above prepared extract was added with dil. HCl till the effervescence ceases. Then 2ml of dil.silver nitrate solution was added.	No cloudy appearance.	Absence of Chloride
3.	**Test For Phosphate:** 2ml of the extract was treated with 2ml of dil.ammonium molybdate solution and 2ml of con.HNo3.	Yellow appearance present	Absence of Phosphate

4 .	**Test For Carbonate:** 2ml of the extract was treated with 2ml dil. Magnesium sulphate solution	No Cloudy appearance.	Absence of carbonate
5.	**Test For Nitrate:** 1gm of the substance was heated with copper turning and concentrated H2So4 and viewed the test tube vertically down.	No Brown gas evolved.	Absence of Nitrate
6.	**Test For Sulphide:** 1gm of the substance was treated with 2ml of con. HCL	No Rotten Egg Smelling gas.	Absence of Sulphide
7.	**Test For Fluoride & Oxalate:** 2ml of extract was added with 2ml of dil. Acetic acid and 2ml dil.calcium chloride solution and heated.	No Cloudy appearance	Absence of fluoride and oxalate
8.	**Test For Nitrite:** 3drops of the extract was placed on a filter paper, on that-2 drops of dil.acetic acid and 2 drops of dil.Benzidine solution was placed.	No Characteristic changes	Absence of Nitrite
9.	**Test For Borate:** 2 Pinches(50mg) of the substance was made into paste by using dil.sulphuric acid and alcohol (95%) and introduced into the blue flame.	No Bluish green colour flame.	Absence of borate

	II. Test For Basic Radicals		
1.	**Test For Lead:** 2ml of the extract was added with 2ml of dil.potassium iodine solution.	No yellow Precipitate obtained.	Absence of Lead
2.	**Test For Copper:** One pinch (50mg) of substance was made into paste with con.HCl in a watch glass and introduced into the non-luminuous part of the flame.	No Blue colour flame No Blue colour precipitate formed.	Absence of copper
3.	**Test For Aluminium:** To the 2ml of extract, dil.sodium hydroxide was added in 5 drops to excess.	NoYellow colour appeared.	Absence of aluminium
4.	**Test For Iron:** a. To the 2ml of extract,2ml of dil.ammonium solution was added. b. To the 2ml of extract 2ml thiocyanate solution and 2ml of con HNo3 was added	blood red colour appeared.	presence of Iron
5.	**Test For Zinc:** To 2ml of the extract, dil.sodium hydroxide solution was added in 5 drops to excess and dil.ammonium chloride was added.	No White precipitate was formed	Absence of Zinc

6.	**Test For Calcium:** 2ml of the extract was added with 2ml of 4% dil.ammonium oxalate solution	No Cloudy appearance and white precipitate was obtained	Presence of calcium
7.	**Test For Magnesium:** To 2ml of extract dil.sodium hydroxide solution was added in drops to excess.	White precipitate was obtained	Absence of Magnesium
8.	**Test For Ammonium:** To 2ml of extract 1 ml of Nessler's reagent and excess of dil.sodium hydroxide solution are added.	No Brown colour appeared	Absence of ammonium
9.	**Test For Potassium:** A pinch(25mg) of substance was treated of with 2ml of dil.sodium nitrite solution and then treated with 2ml of dil.cobalt nitrate in 30% dil.glacial acetic acid.	No Yellowish precipitate was obtained.	Absence of Potassium
10.	**Test For Sodium:** 2 pinches(50mg) of the substance was made into paste by using HCl and introduced into the blue flame of Bunsen burner.	yellow colour flame appeared	Absence of sodium
11.	**Test For Mercury:** 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No yellow precipitate was obtained	Absence of mercury

12.	**Test For Arsenic:** 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No brownwish red precipitate was obtained	Absence of arsenic
	III. Miscellaneous		
1.	**Test For Starch:** 2ml of extract was treated with weak dil.iodine solution	No blue colour developed	Absence of starch
2.	**Test For Reducing Sugar:** 5ml of Benedict's qualitative solution was taken in a test tube and allowed to boil for 2 minutes and added 8 to 10 drops of the extract and again boil it for 2 minutes. The colour changes are noted.	Brick red colour not developed	Absence of reducing sugar
3.	**Test For The Alkaloids:** a) 2ml of the extract was treated with 2ml of dil.potassium iodide solution. b) 2ml of the extract was treated with 2ml of dil.picric acid. c) 2ml of the extract was treated with 2ml of dil.phosphotungstic acid.	. Yellow colour developed	- Presence of Alkaloids
4.	**Test For Tannic Acid:** 2ml of extract was treated with 2ml of dil.ferric chloride solution	No black precipitate was obtained	Absence of Tannic acid

5.	**Test For Unsaturated Compound:** To the 2ml of extract 2ml of dil.Potassium permanganate solution was added.	Potassium permanganate was not decolourised	Absence of unsaturated compound
6.	**Test For Amino Acid:** 2 drops of the extract was placed on a filter paper and dried well. 20ml of Biurette reagent was added.	violet colour developed	Presence of amino acids
7.	**Test For Type Of Compound:** 2ml of the extract was treated with 2 ml of dil.ferric chloride solution.	No green colour developed No red colour developed No violet colour developed No blue colour developed	Absence of oxy quinole pinephrine and pyro catechol Anti pyrine, Aliphatic amino acids and meconic acid are absent Apomorphine salicylate and Resorcinol are absent Morphine, Phenol cresol and hydro uinone are absent

TOXICITY STUDY

ACUTE AND SUB ACUTE TOXICITY STUDY OF VALENDRAPHOLA CHOORANAM IN RODENTS

Animals:

Mice of either sex weighing 25-30g and rats weighing 210-240g were obtained from the animal house of Vels University. The animals were used with the approval of the Institute Animal Ethics Committee and obtained from Vels University, Chennai. They were fed with a balanced standard pellet diet and maintained under standard laboratory conditions, providing 24-28^{0}C temperature, standard light cycle (12 h light, 12 h dark) and water ad libitum. Animals were kept in cages with raised floors of wide mesh to prevent coprophagy. Animal welfare guidelines were observed during the maintenance period and experimentation. The rats were randomly assigned to control and different treatment groups, six animals per group. (Approval number: XIII/VELS/PCOL /38/2000/CPCSEA/IAEC/08.08.2012). The animals were acclimatized for one week under laboratory conditions.

ACUTE TOXICITY STUDY-OECD 425 GUIDELINES

Acute oral toxicity test for the Valendraphola Chooranam was carried out as per OECD Guidelines 425. As with other sequential test designs, care was taken to ensure that animals are available in the appropriate size and age range for the entire study. The test substance was administered in a single dose by gavage using a stomach tube or a suitable intubation cannula. The fasted body weight of each animal was determined and the dose was calculated according to the body weight. After the substance has been administered, food was withheld for a further 2 hours in mice. The animals were observed continuously for the first

4 h and then each hour for the next 24 h and at 6 hourly intervals for the following 48 h after administering of the test drug, to observe any death or changes in general behaviour and other physiological activities. Single animals are dosed in sequence usually at 48 h intervals. However, the time interval between dosing was determined by the onset, duration, and severity of toxic signs. Treatment of an animal at the next dose was delayed until one was confident of survival of the previously dosed animal.

Observation of toxicity signs:

General behaviour, respiratory pattern, cardiovascular signs, motor activities, reflexes, change in skin and fur, mortality and the body weight changes were monitored daily. The time of onset, intensity, and duration of these signs, if any, was recorded.

ACUTE TOXICITY STUDY

Acute oral toxicity test for the Valendraphola Chooranam was carried out as per OECD Guidelines 425. Animals were observed individually after dosing at least once during the first 30 minutes, periodically during the first 24 hours, with special attention given during the first 4 hours, and daily thereafter, for a total of 14 days, except where they need to be removed from the study and humanly killed for animal welfare reasons or are found dead.

All observations are systematically recorded and Observations include changes in skin and fur, eyes and mucous membranes, and also respiratory, circulatory, autonomic and central nervous systems, and somatomotor activity and behaviour pattern. Attention was directed to observations of tremors, convulsions, salivation, diarrhoea, lethargy, sleep and coma. The principles and criteria summarised in the Humane Endpoints Guidance Document taken into consideration. Animals found in moribund condition and animals showing severe pain or enduring signs of severe distress was humanely killed. When animals were killed for human reasons or found dead, the time of death was recorded.

SUB-ACUTE TOXICITY

In a 28-days sub acute toxicity study, twenty four rats of either sex (3+3) were divided into four groups of 6 rats each. Group I that served as normal control was administered with distilled water (p.o.) while groups II, III and IV were administered daily with the Valendraphola Chooranam (p.o.) for 28 days at a dose of 0.1, 0.25 and 0.5 g/kg respectively. The animals were then observed daily for gross behavioural changes and any other signs of subacute toxicity. The weight of each rat was recorded on day 0 and weekly throughout the course of the study, food and water consumption per rat was calculated. At the end of the 28 days they were fasted overnight, each animal was anaesthetized with diethylether, following which they were then dissected and blood samples were obtained by cardiac puncture into heparinised tubes. The blood sample

collected from each rat was centrifuged with 3000 X g at 4^{o}C for 10 min to separate the serum and used for the biochemical assays.

Hematological and blood biochemical analyses:

At the end of the study, all animals were kept fasted for 16-18 h and then anesthetized with anesthetic ether on the 28th day. Blood samples for hematological and blood chemical analyses were taken from retro orbital vein. Heparinized blood samples were taken for determining complete blood count (white blood cell count, differential white blood cell count, platelet count, red blood cell count, hematocrit, and hemoglobin) by semiautomated hematology analyzer. The serum from non-heparinized blood was carefully collected for blood chemistry and enzyme analysis (glucose, blood urea nitrogen (BUN), creatinine, total protein, albumin, total and direct bilirubins, serum glutamate-oxaloacetate transaminase (SGOT), serum glutamate pyruvate transaminase (SGPT), and alkaline phosphatase (ALP)) were automatically determined using autoanalyzer.

Necropsy:

All rats were sacrificed after the blood collection. The positions, shapes, sizes and colours of internal organs were evaluated. The Spleen, Testes, Pancreas, Lung, Liver, Brain, Heart, Stomach, Intestine, Bone, Ovary, and Kidney tissues were excised from all rats to visually detect gross lesions, and weighed to determine relative organs' weights and preserved in 10% neutral formalin for histopathological assessment. The tissues were embedded in paraffin, and then sectioned, stained with haematoxylin and eosin and were examined microscopically.

STATISTICAL ANALYSIS

Values were represented as mean ± SEM. Data were analysed using one-way analysis of variance (ANOVA) and group means were compared using the Tukey-Kramer Multiple Comparisms Test using Graph Pad Instat-V3 software. P values < 0.05 were considered significant. (Trial drug I, Table 4 – Table 11)

RESULTS

In the acute toxicity study at the dose level of 5 g/kg moderate toxic symptoms like alertness, grooming, touch response, writhing and hypnosis was observed. Hence the

next lower dose was tried and confirmed the non toxic response to test drug Valendraphola Chooranam. Hence for the further study the one tenth, one fifth and one twentieth of the tolerable dose was selected for the further sub acute toxicity evaluation.

In the sub acute toxicity study, Animals were not shown any significant toxic clinical signs during the dosing period of 28 days. All animals from control and all the treated dose groups survived throughout the dosing period of 28 days.

Results of body weight determination of animals of control and different dose groups exhibited comparable body weight gain throughout the dosing period of 28 days. During dosing period, the quantity of food consumed by animals from different dose groups was found to be comparable and normal with that of control animals.

Ophthalmoscopic examination of animals in control and test product treated groups did not reveal any major and remarkable abnormality. Urine analysis data of control group and treated group of animals determined in week 4 did not reveal any abnormalities.

Comparison of organ weights of treated animals with respective control animals on day 28 kidney weight was slightly increased at middle dose treated group and was found to be comparable.

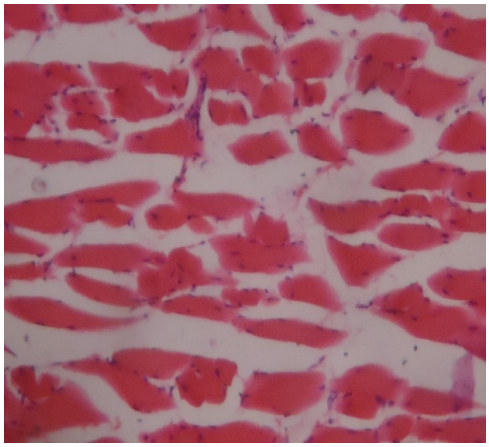
Gross pathological examination of animals in control as well as the treated groups did not reveal any abnormalities. The increase or decrease in the values obtained was within normal biological and laboratory limits.

An increase in total RBC count and Hb was obtained for animals in the dose group of 250 and 500 mg/kg ($P<0.01$). Results of Biochemical investigations conducted on days 28 no significant changes in the values of different parameters studied when compared with control; however, the values obtained were within normal biological and laboratory limits.

CONCLUSION

Based on these findings, no toxic effect was observed upto 500mg/kg of Valendraphola Chooranam via oral route over a period of 28 days. So, it can be concluded that the Valendraphola Chooranam can be prescribed for therapeutic use in human with the dosage recommendations of upto 500 mg/kg. body weight p.o.

HISTOPATHOLOGY SLIDES



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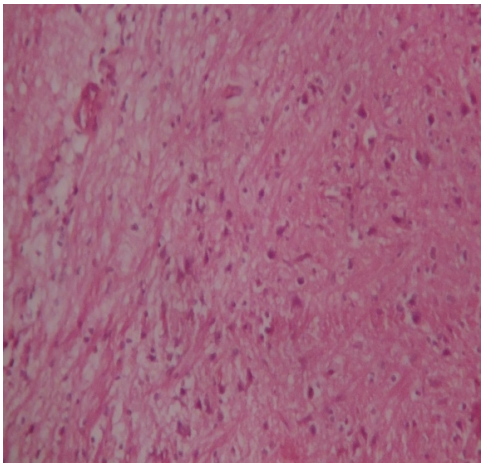

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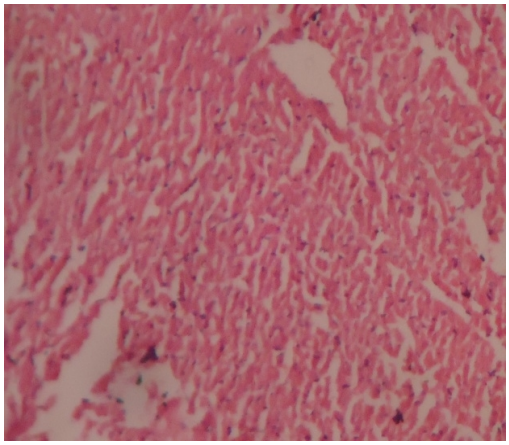

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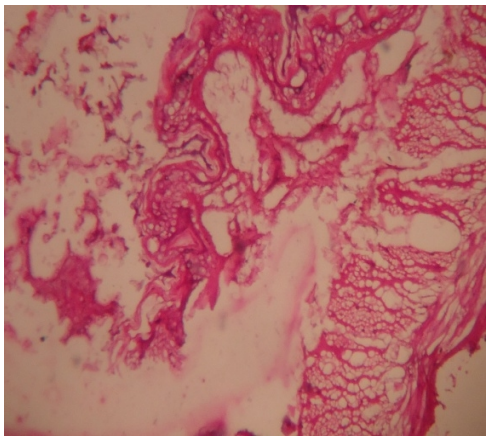
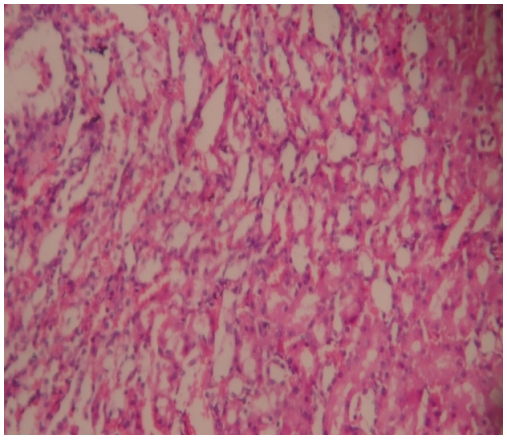
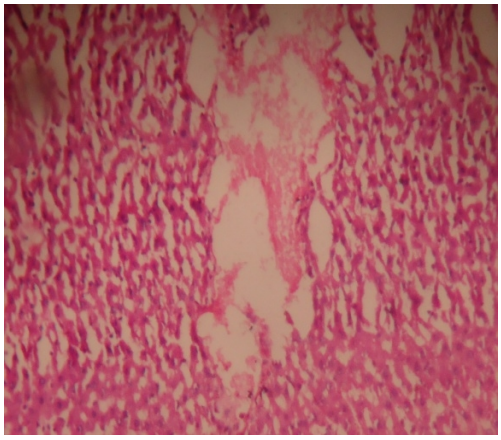


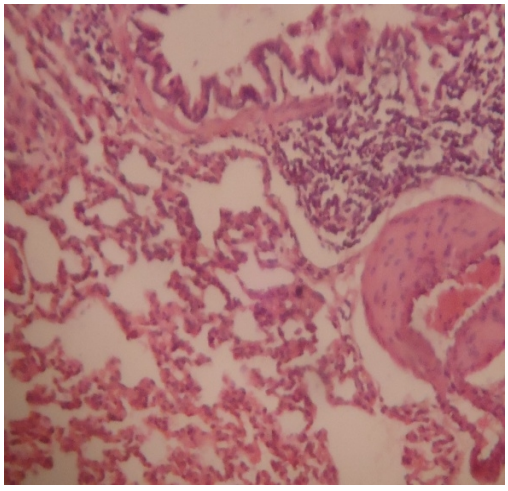
INTESTINE



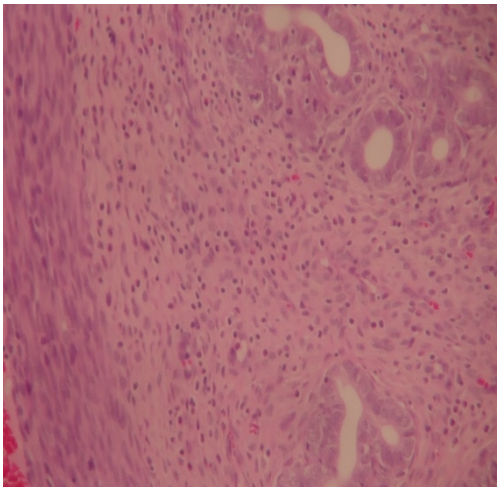
KIDNEY



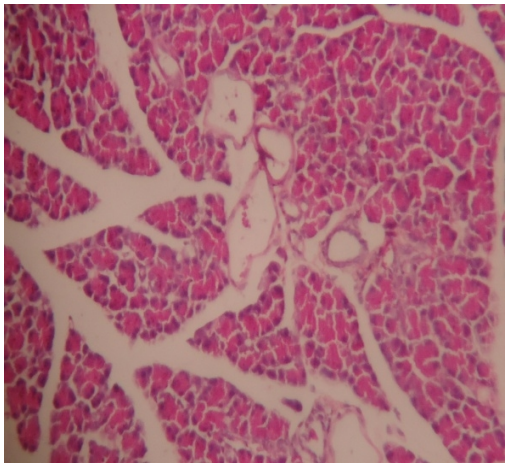
LIVER



LUNG



OVARY



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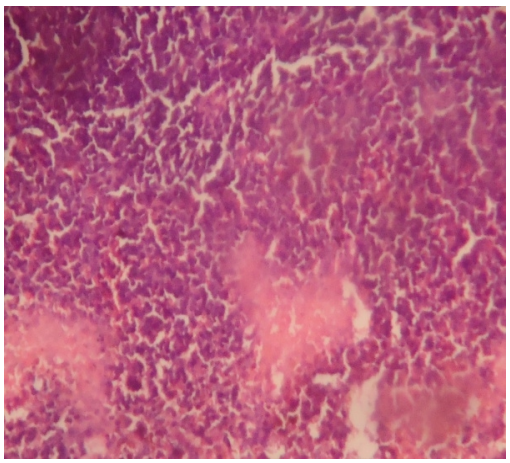
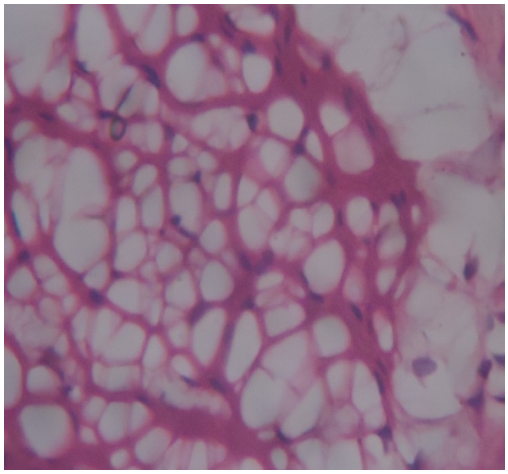
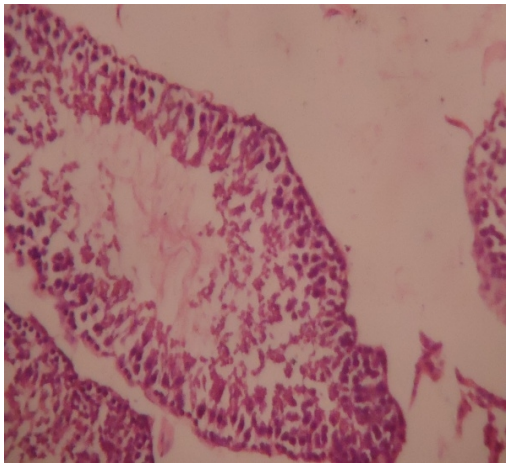


SPLEEN



STOMACH



TESTIS

PHARMACOLOGICAL STUDY

HAEMATINIC ACTIVITY OF VALENDRAPHOLA CHOORANAM

MATERIALS AND METHODS

Drug material

The suspension of Valendraphola Chooranam was stored in a refrigerator until use. The suspension was further diluted with 2% CMC to get 200mg/ml stock solution and used in this study.

Animals

Male albino rats (150-180g) were used for the study. The rats were housed in wire-mesh cages with a 12 h light/dark cycle. They had continuous access to fod and water during the entire period of experimentation.

(APPROVAL NUMBER: XIII/VELS/PCOL/38/2000/CPCSEA/IAEC/08.08.2012)

EVALUATION OF HAEMATINIC ACTIVITY

Six rats were kept as normal control group (Group 1), while 24 rats were made anaemic by oral intubations of phenylhydrazine (10 mg/kg body weight) daily for 8 days. Rats that developed anaemia with haemoglobin concentration <14 g/dl were recruited for the study. Anaemic rats were randomly divided into 5 groups (2 to 6) and treated as follows:

Group 1: received distilled water (1 ml) daily (normal control)

Group 2: received 2% CMC (1 ml) daily (anaemic control)

Group 3: received oral single dose of the Valendraphola Chooranam 100 mg/kg body weight/day

Group 4: received oral single dose of the Valendraphola Chooranam 200 mg/kg

Group 5: received oral single dose of the Valendraphola Chooranam 400 mg/kg

Group 6: received oral single dose of the heamatinic syrup 2ml/kg body weight/day.

The treatment was continued for 2 weeks.

Haematological investigation

Blood collected from the retro orbital vein of experimental animals after an overnight fast and after 1 and 2 weeks of treatment with Valendraphola Chooranam, was used for the determination of red blood cell count (RBC), haemoglobin (Hb) concentration and packed cell volume (PCV). The mean cell volume (MCV), mean cell haemoglobin (MCH) and the mean cell haemoglobin concentration (MCHC) were calculated.

STATISTICAL ANALYSIS

Experimental data was analysed using analysis of variance (ANOVA) and Dunnet's 't' test to determine significant differences between means. The statistical analysis system (INSTAT-V3) package was used for this analysis. (Trial drug I, Table 12 – Table 14, Chart 1 - 3)

RESULTS AND DISCUSSION

This study aimed to evaluate the effect of Valendraphola Chooranam on the haemolytic anaemia induced by phenylhydrazine in albino rats. There was a significant ($p<0.05$) increase in the mean Hb concentration recorded for rats in the test group 200mg/kg of Valendraphola Chooranam relative to that observed in the control group (15.64 ± 0.59 g/dl).

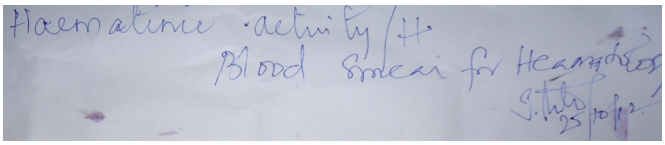
After one week of treatment, the mean total red blood cell count of rats in each of the Valendraphola Chooranam test groups (5.16±0.24, 5.28±0.18, 5.11±0.21X 10^6/µL) were observed compared with those in the control group (4.12±0.28X 10^6/µL), but these differences were not significant ($P>0.05$). There was an increase in the PCV of rats in all the Valendraphola Chooranam treatment groups relative to that of the rats in the control group. The increase was significant ($p < 0.05$) for rats in high dose group (49.46 ± 2.48%), compared with control group. There was a slight increase observed in the mean MCV values of rats in all the drug treated groups but it was statistically not significant compared to control group.

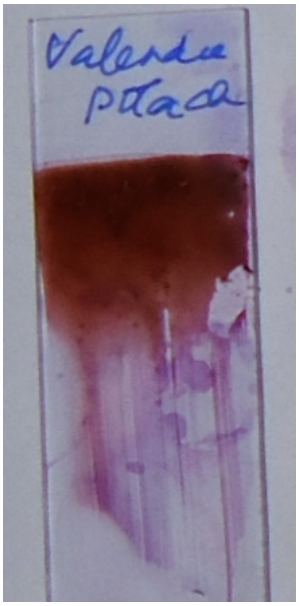
Rats in low dosed test group were observed to have decrease mean MCH values ($p<0.05$). There was a non-significant ($p>0.05$) decrease in the mean MCHC values of rats in all the test groups.

After 14 days of Valendraphola Chooranam treatment significantly increased almost all the parameters towards normal. Hb level was 17.10±0.68, 17.15±0.56, 18.56±0.54g/dl respectively in 100, 200 and 400 mg/kg Valendraphola Chooranam treated groups. In the same manner, the RBC count was rapidly increased to 4.81±0.34 at Valendraphola Chooranam mid dose treated group ($P<0.05$). The mean RBC and Hb concentration of rats were increased, especially for rats administered the higher doses. Mean MCV values were also increased, while MCH and MCHC values decreased.

CONCLUSION

Iron deficiency adversely affects the cognitive performance, behaviour, and physical growth of infants, preschool and school-aged children. The ideal antianaemic regimen should be safe, cheap, well tolerated by children and able to achieve a high cure rate. Iron preparations available used in treatment of iron deficiency anaemia has much adverse effect. In this study, the test drug Valendraphola Chooranam justifies and supports the traditional use in the treatment of anaemia.





DISEASE ASPECT

SIDDHA ASPECT

வெளுப்பு நோய்[13]

வேறுபெயர் : வெண்மை நோய், பாண்டு.

இயல்பு :

இயற்கை நிறம்மாறி, உடல் வெளுத்து, கண்ணையும் நகக்கண்ணையும் நீக்கிப் பார்க்கின் குருதியின்றி வெளுத்திருக்கும்.

நோய் தோன்றும் வழி :

குருதியின் வன்மைமையைக் குறைக்கக் கூடிய உப்பு, புளிப்புள்ள பொருட்களை மிகுதியாகக் கொள்வதாலும், சுரம், பேதி, வாந்தி, கீல்வாயு முதலிய நோய்களுக்கு உட்படுதலாலும், குருதியை அளவு கடந்து வெளியாக்கும் பெரும்பாடு, குருதியழல் நோய், குருதிக் கழிச்சல், முளைநோய் (மூலம்), குருதிவாந்தி, முதலியவை ஏற்படுதலாலும் வெட்டுப் பட்டு மிகுதியாகக் குருதி வெளிப்படுதலாலும் இந்நோய் உண்டாகும். அன்றியும் நச்சுத்தன்மையுடைய மருந்துகளை நாளளவுக்கு மிஞ்சி உண்பதாலும், புகையிலை, வெற்றிலைப் பாக்கு, மண், சாம்பல், திருநீறு, கற்பூரம் முதலியவைகளை அடிக்கடி கொள்வதாலும் இந்நோய் வரும்.

முற்குறிகள் :

உணவு முதலிய வேறுபாடுகளால், தீக்குற்றம் மிகுந்து, குருதியின் நிறத்தையும், எடையையும் கெடுத்து, உடற்கு வேண்டிய ஊட்டத்தையும் கொடாமல், உடலை வெளுக்கச் செய்யும். சிறிது தொலைவு நடக்கினும் கால் ஓய்ந்து போதல், பெருமூச்சு வாங்கல், உணவில் விருப்பமின்மை, வாய்குமடட்ல், தலைசுற்றல், கண் இருளல் அடிக்கடி மயக்கமாதல், மார்பு துடித்தல், உடல் இளைத்தல் உண்டாகும்.

நோய் எண்:

குற்றத்தால் வருவன நான்கும், நஞ்சால் வருவன ஒன்றும் கூடி ஐந்தாகும்.

1. வளி (வாதப்) பாண்டு

2. தீ (பித்தப்) பாண்டு

3. ஐய (கபப்) பாண்டு

4. முக்குற்றம் (திரிதோஷ) பாண்டு

5. நஞ்சு (விடப்) பாண்டு,

மண்ணுன் வெளுப்பு (பாண்டு) நோய் ஒன்றுளது என்றும் சிலர் கூறுவர்.

வளி வெளுப்பு நோய் :

நீர்வேட்கை, பசி அற்று, குருதி நாளங்கள் கறுப்பாகி, பரபரத்துக் காணும். கண் சிவக்கும், உடல் வெளுத்து வீங்கும். உணவிற் சுவை யறியாமை, வயிறு பொருமல் உண்டாகும்.

அழல் வெளுப்பு நோய் :

“வாமென்ற மேனியெலா மஞ்ச ளித்து
மகாவெளுப்பு உண்டாகி மந்தக் கண்ணாந்
தாமென்ற தாகமொடு மூர்ச்சை யாகும்
தனிவாயில் மிளகுபோற் றானு றைக்கும்
நேமென்ற நெஞ்சுமுள் தானு முண்டாய்
நெருக்கியே மூச்சுமுட் டதுவே யாகுங்
கோமென்ற கிறுகிறுத்து வாய்கைப் பாகுங்
கிளர்பித்த பாண்டுவெனக் கூறலாமே”

நா, கை, கால்கள் வெளுத்து, கண் பார்வை மங்கும். நீர்வேட்கையோடு மயக்கம், நெஞ்சை இறுக்கிப் பிடித்தது போல் மூச்சு முட்டுவதோடு தலை கிறுகிறுக்கும்.

குளிர்ச்சி தரும் பொருள்களில் விருப்பு, வாய் புண்ணாதல்.

ஐய வெளுப்பு நோய் :

தோல் மிக வெளுத்து நரம்புகள் புடைக்கும். நா உப்பு கைத்தல், வாந்தியாதல், மயக்கமடைதல், இடுப்பு நோதல், மேல்மூச்சு வருதல், உடல் வன்மை குறைந்து மார்பு துடித்தல், அடிக்கடி உடல் முழுமையும் ஊதும்.

நஞ்சு வெளுப்பு நோய் :

உடல் வெளுக்கும். நீர் வேட்கை, சுவையின்மை, வாந்தி, விக்கல், பெருமூச்செரிதல், உடல் முழுமையும் வீங்கும்.

முக்குற்ற வெளுப்பு நோய் :

மேல்மூச்சு வருதல், உடல் வன்மை குறைந்து மார்பு துடித்தல், அடிக்கடி உடல் முழுமையும் ஊதும்.

மண்ணுன் வெளுப்பு நோய் :

இஃது சிறு குழந்தைகளும், சிறுவயதினரும், கருவுற்றவரும், மண், சாம்பல், செங்கல், திருநீறு, கருப்பூரம் இவைகளின் மீது தனித்த இச்சை கொண்டு, அளவு கடந்து உண்பதால் காணும் நோயாகும். செரியாமை, வாந்தி, உடல் மெலிந்து, குருதிவற்றி, வெளுத்து வீங்கி மார்பு துடிக்கும்.

குற்றம் முதலிய வேறுபாடுகள் :

அழல் (இரஞ்சித பித்தமும்) மெலிந்து, நிறத்திலும் எடையிலும் குறைந்து, தீக்குற்றத்தைப் பெருக்கும். அதனளவாக மற்றைய குற்றங்களும் தன்னிலையில் திரிந்து, பரவுகாலின் வன்மையைக் கெடுத்து நோயை உண்டாக்கும்.

நாடி :

“தானமுள்ள சேத்துமந்தானிளிகில் .....

.....

..... பாண்டுரோகம்”

“இடமான சேத்துமத்திற் பித்தநாடி
எழுந்தணுகில் பாண்டாகும்”
“உண்டாயோ சேத்துமத்தில் வாதநாடி
கலந்திடுமேல் பாண்டு பிறக்குந்தானே”
“வாதத்தில் சீதஞ்சேர்ந்தால் பாண்டுண்டாமே”

MODERN ASPECT

ANAEMIA

Anaemia is defined as a reduction of haemoglobin levels below the normal values for the different age and sex groups[14].

The normal values of haemoglobin for different age groups accepted by WHO are as follows[14]:

Children (6 months to 6 years)	:11 g/dL and above
(6 years to 14 years)	:12 g/dL and above
Adult males	:13 g/dL and above
Adult females (non pregnant)	:12 g/dL and above
Adult females (pregnant)	:11 g/dL and above

In full health there is no racial variation in haemoglobin. In India any haemoglobin level below 12 g/dL in adult males and 11.5 g/dL in adult females should be diagnosed as anaemia and investigated. In general, reduction of haemoglobin is associated with a fall in erythrocyte count and Packed cell volume(PCV)[14].

Causes of Anaemia[15]:

Decreased or ineffective marrow production

- Inadequate iron, B12 or folate, trace elements(zinc, cobalt)
- Hypoplasia of bone marrow
- Infiltration by malignant cells.

Peripheral causes(increased RBC destruction or loss)

- Blood loss
- Haemolysis
- Hypersplenism

Classification of anaemia[15]:

Classification of anaemia based on RBC Morphology:

- Normocytic anaemia
- Macrocytic anaemia
- Microcytic anaemia

IRON DEFICIENCY ANEMIA[14]:

Microcytic hypochromic anaemia occurring in India is mostly due to iron deficiency. In India and other developing countries, Iron deficiency anaemia far out numbers all the other types of anaemias put together, as it constitutes 90-95% of the total. Iron deficiency anaemia is one of the most widespread diseases all over the world.

Iron deficiency is prevalent in 30-50% of the adolescent and young adult women due to their unsatisfactory food habits and moderate or heavy blood loss during menstruation.

Even though iron deficiency is mainly caused by inadequate iron intake in food, Iron deficiency anaemia is not exclusively a disease of the poor. Food fadism and other diseases which cause blood loss account for the majority of anaemia cases occurring in the rich.

Causes of Iron deficiency anemia[16]:

Defective Intake:

In children, psychiatric patients, patients having anorexia due to any cause.

Defective absorption:

Gastrectomy

Gastrojejunostomy

Excessive demand:

Growing children

Females during reproductive period of life.

Excessive loss:

Bleeding piles

Menorrhagia in females

Recurrent blood donation

Long standing haematuria

Drugs including Aspirin.

Clinical features[16]

Symptoms:

Weakness

Fatigue

Lassitude

Lustreless hair

Palpitation

Breathlessness

Giddiness

Tingling

Numbness

Insomnia

Lack of concentration

Angular stomatitis

Signs:

Pallor

Dry skin

White sclera

Koilonychia

Investigations:

Hb

HCT/PCV

MCV

MCH

MCHC

Peripheral smear study

Serum iron

TIBC

Serum ferritin.

CLINICAL STUDY

The study was conducted on patients with Pandu [Anaemia] patients satisfying the inclusion criteria.

The study was conducted at the OPD/IPD of Ayothidoss Pandithar Hospital of the National Institute of Siddha, Tambaram sanatorium, Chennai-47.

Sample size:

The sample size was 20 patients.

SUBJECT SELECTION:

Inclusion criteria:

Age : 20-60 yrs

Sex : Both male and female

Weight : 35-85 kgs

Patient having symptoms of

- Fatigue
- Lassitude
- Dyspnoea on exertion
- Giddiness
- Headache
- Insomnia
- Palpitation
- Poor appetite

Any of the above 4 clinical symptoms

Patient who are willing to provide blood sample for lab investigation.

Patient who are willing to attend OPD once in 7 days.

Patient who are willing to be admitted in the hospital for 30 days.

Patient willing to sign the informed consent stating that he/she will conscientiously stick to the treatment during 30 days but can opt out of the trial of his/her own conscious discretion.

Exclusion criteria:

Pregnancy and lactation

Cardiac diseases

Known case of Cirrhosis and Chronic Renal Failure

Patient receiving Anti-tuberculosis drugs

Hypo/hyperthyroidism

Chronic blood loss

Any other serious illness

Withdrawal criteria:

Development of any adverse reaction

Occurrence of any other serious illness

Non co-operation of the patient

TRIAL DRUG AND DURATION

Drug **:**Valendraphola Chooranam -1 gm, bid with luke warm water, after food.

Duration of the treatment: 30 days.

CONDUCT OF THE STUDY:

Pandu patients satisfying inclusion and exclusion criteria were admitted to the trial. Informed consent was obtained from the patients. Routine investigations like Blood test, peripheral smear study, urine test were carried out before and after the trial treatment. For in patients the drug was administered daily. For out patients the trial drug was issued for seven days course. They were advised to visit the OPD once in 7 days. At each visit they were clinically assessed.

CLINICAL OBSERVATION:

For the clinical study of "Valendraphola Chooranam" on Pandu, 20 patients were selected.

Among 20 patients,17(85 **%)** were female, 3(15%) were male.

According to age wise distribution 20% were in 20-30years, 40% were in 31-40years, 25% were in 41-50 years and 15% were in 50-60 years.

Among 20 patients, All were affected from pallor, 17 patients were affected from breathlessness, 18 patients were affected from tiredness, 12 patients were affected from giddiness, 18 patients were affected from anorexia, 13 patients were affected from pica and 16 patients were affected from palpitations.

From the clinical study 10% of patients relieved from pallor, 76.47% of patients relieved from breathlessness, 83.33% of patients relieved from tiredness, 83.33% of patients relieved from giddiness and 83.33% of patients relieved from anorexia, 69.23% patients were relieved from pica and 81.25% patients were relieved from palpitations and no adverse effects were observed during trial period**.**

16 (80%) Patients had significantly improved in the Hb levels after treatment.

The following investigations were done before and at the end of the treatment.

- Blood sample (Hb, TC, DC, ESR, HCT/PCV, MCV, MCH, MCHC, and smear study, Sugar (F, PP), AEC, T.cholesterol, RFT and LFT)
- Urine test (Albumin, Sugar, Deposits)
- Motion test (ova, cyst, albumin)

DISCUSSION

The drug Valendraphola Chooranam was selected to evaluate the Haematinic activity in the management of Pandu (Anaemia).

The literary evidence from Gunapadam mooligai vaguppu strongly supports the Haematinic activity of the drug.

Bio-chemical analysis:

The biochemical analysis of the drug reveals the presence of iron, calcium, alkaloid and amino acids.

Iron[17]:

It is a component of haem which is required for the formation of haemoglobin and hence it compensates the blood loss. It helps in the transport of Oxygen and may be useful in preventing hypoxia, which is one of the cause for anemia.

Hence the iron increases the haemoglobin concentration, thereby it reduces the anaemia.

Calcium[18]:

Calcium plays a very important role in the body. It is necessary for normal functioning of nerves, cells, muscle, and bone. Hence it is considered as a nutritive supplement for treating Anaemia.

Toxicological studies:

Based on Sub acute toxicity Study, no toxic effect was observed upto 500 mg/kg of Valendraphola Chooranam via oral route over a period of 28 days. So, it can be concluded that the Valendraphola Chooranam can be prescribed for therapeutic use in human with the dosage recommendations of upto 500 mg/kg. body weight p.o.

Pharmacological studies:

In this study, the test drug Valendraphola Chooranam justifies and supports the haematinic activity and traditional use in the treatment of anaemia.

Clinical observation:

10% of patients relieved from pallor, 76.47% of patients relieved from breathlessness, 83.33% of patients relieved from tiredness, 83.33% of patients relieved from giddiness and 83.33% of patients relieved from anorexia, 69.23% patients were relieved from pica and 81.25% patients were relieved from palpitations and no adverse effects were observed during trial period**.**

16 (80%) Patients had significantly improved in the Hb levels after treatment.

Bio-statistics:

Statistically, the paired 't' test shows statistical significance for the Hb levels and associated symptoms before and after the treatment.($p<0.0001$)

Siddha Aspect[19]:

சுவை : கைப்பு

தன்மை : வெப்பம்

பிரிவு : கார்ப்பு

கைப்பு சுவை அரோசகத்தை நீக்கும். பித்தம், கபம் விகற்பத்தினை சாந்தி செய்யும்.

வெப்ப வீரியமானது கபத்தை நீக்கும், தலைச்சுற்றல், நாவறட்சி, தேகவாட்டம், விரைவில் செரிப்பித்தல் ஆகிய காரியங்களை செய்யும்.

கார்ப்பு சுவையானது அசீரணம், வயிற்றுப் பொருமல், சோகை ஆகியவற்றை நீக்கும், தீபனத்தையும், செரிப்பையும், நற்சுவையையும் உண்டாக்கி, கெட்ட மலத்தைக் கழிப்பித்து, கபத்தால் உண்டான கேடுகளை நீக்கும்.

எனவே பித்தம், கபம், இவற்றின் குற்றங்களால் எற்படும் பாண்டு நோயினை கைப்பு, வெப்பம், கார்ப்பு வீரியமுடைய மூலிகைகளை கொண்டு மருத்துவம் செய்ய நற்பலனை அளிக்கும்[7].

SUMMARY

The drug Valendraphola Chooranam was selected to evaluate the Haematinic activity in the management of Pandu (Anemia).

The literary evidence from Gunapadam mooligai vaguppu strongly supports the Haematinic activity of the drug.

The qualitative and quantitative analysis were done at Biochemistry lab, National Institute of Siddha and Sri Ramachandra University, Chennai respectively. The biochemical analysis of the drug reveals the presence of iron, calcium, alkaloid and amino acids. The result ensures the Haematinic activity of the Valendraphola Chooranam was due to the presence of active phytoconstituents of the drug.

High Performance Thin Layer Chromatography was done at Sri Ramachandra University, Chennai.

The pre-clinical evaluation (acute & repeated oral toxicity study) of the drug was carried out as per OECD guideline in Vels college of Pharmacy, Chennai. The result shows the safety of the drug for human administration.

The Preclinical Pharmacological study was carried out in animal model in Vels college of Pharmacy, Chennai. The result shows that the drug has significant Haematinic effect.

As per the Siddha literature and modern science reviews and research articles, the trial drug has potent Haematinic effect.

20 Patients were recruited for clinical trial. The trial drug Valendraphola chooranam at the dose of 1 gm, b.i.d was given to the patient for 7 days and patients were asked to visit op once in 7 days for 30 days. Clinical assessment and prognosis was noted at each visit.

From the clinical study, after the course of treatment, 16 (80%) of the patients had significant increase in the Hb levels after treatment.

10% of patients relieved from pallor, 76.47% of patients relieved from breathlessness, 83.33% of patients relieved from tiredness, 83.33% of patients relieved

from giddiness and 83.33% of patients relieved from anorexia, 69.23% patients were relieved from pica and 81.25% patients were relieved from palpitations and no adverse effects were observed during the trial period**.**

Statistically, the paired 't' test shows statistical significance for the Hb levels and associated symptoms before and after the treatment.($p<0.0001$)

The drug Valendraphola Chooranam has

- Haematinic Activity
- No side effects
- No undoing effects
- Encouraging clinical results.

From the clinical study and the statistical analysis, it was proved that the drug Valendraphola Chooranam was statistically significant on Haematinic activity in the management of Pandu (Anemia).

CONCLUSION

The literature evidence of the plant shows that it has Haematinic activity.

The safety studies (acute toxicity and repeated oral toxicity studies) conducted revealed that the trial drug Valendraphola Chooranam is safe. There were no abnormalities found in blood investigation and histo-pathological examination. Hence it can be reasonably assumed that the drug is safe for human use.

The pharmacological study conducted in animal model shows significant Haematinic activity.

Clinical study reveals the therapeutic efficacy of the trial drug by showing, increasing in Hb levels significantly. There was improvement in clinical symptoms after treatment.

There were no adverse reactions complained during the clinical trial.

Hence, the drug VALENDRAPHOLA CHOORANAM can be used in the management of Pandu (Anaemia).

INTRODUCTION

Bronchial asthma is one of the major health issues in many developing countries, causing great concern and stress among the victims[20]. Asthma is a disease which dates back atleast to the time of Hippocrates, who noted a condition of "deep and heavy breathing". The Greeks had labelled this condition as 'Asthma', the meaning of which was panting[21].

Most of the individuals develop severe exacerbation of asthma by several triggering agents including increasing air pollution, urbanization, stress, deforestation. Home factors that can lead to exacerbation include dust, house mites, animal danders, cockroach allergens. The situation is complicated by poor access to medical services and poor health education among the affected population[22].

In many countries the prevalence of asthma is increasing, particularly in the second decade of life where this disease affects 10-15% of population. There is also a geographical variation with asthma being common in more developed countries. Long term follow up in developing countries suggests that the disease may become more frequent as individuals become more westernised[23].

An estimated 300 million people in the world currently have asthma and there may be additional 100 million persons asthma by the year 2025. It has been reported that there are approximately 15-20 million asthmatics[24]. Asthma is a heterogeneous disease and genetic and environmental factors such as viruses, occupational exposures and allergens contribute to its situation and continuance[25]. Studies of occupational asthma suggest that a high percentage of the work force, perhaps upto 20% may become asthmatic if exposed to potent sensitizers[23].

India contributes to 10% of the global burden of asthma having around 2.4 crore of its population being asthmatic[26].

According to the "The Global Asthma Report 2011", published by the International Union Against Tuberculosis and Lung disease and International study of Asthma and Allergies in childhood: Asthma is the most common chronic disease among children and over 23.5 crore people around the world was affected by it which accounts for 1 in every 250 deaths globally[26].

In Siddha system of medicine, Eraippu noi is compared to Bronchial Asthma. Siddha medicine means medicine that is perfect. Siddha medicine is claimed to revitalize and rejuvenate dysfunctional organs that cause the disease and to maintain the ratio of vatham, pitham and kabam. According to the Siddha medicine, diet and lifestyle play a major role not only in health but also in curing diseases[27].

Immunomodulating agents are useful in the treatment of asthma by inhibiting the antigen-antibody (AG-AB) reaction and thereby inhibiting the release of inflammatory mediators[28].

The preparation "SINGATHI CHOORANAM" is mentioned in the Siddha text "Agasthiyar mani ennum Vaithiya Chinthamani Venba 4000"[29]. The preparation contains Karkadagasingi, Siruthekku, Chukku, Milagu, Thippili, Kadukkai, Thandrikkai, Aamanakku ver, Kandankathari ver, Jatamanjil, Indhuppu. In this Siruthekku, Thippili, Karkadagasingi has Anti-inflammatory effect, Chukku has immunomodulatory effect, Kadukkai has Anti-histaminic activity, Kandankathari ver has Anti-allergic activity.

According to Siddha system of medicine, Eraippu occurs as a result of derangement of kabam[7]. The ingredients of this medicine contains Chukku, Milagu, Thippili, Kadukkai which acts as rejuvenators. Most of the ingredients have Kaarppu suvai, veppa veeriyam and kaarppu pirivu, Kaarppu suvai neutralises the kabam and thereby it may help reducing the exacerbation of attacks.

Hence the author have selected the medicine "SINGATHI CHOORANAM" for treating Eraippu.

AIM:

To evaluate the safety and efficacy of "Singathi Chooranam" for bronchodilator activity in the management of Eraippu [Bronchial Asthma].

OBJECTIVE:

Primary objective:

To evaluate the bronchodilator activity of "Singathi Chooranam" in preclinical studies.

Secondary objective:

Biochemical analysis.

To evaluate the efficacy of "Singathi chooranam" for bronchodilator activity in the management of Eraippu [Bronchial Asthma].

High Performance Thin Layer Chromatography

Atomic Absorption Spectrophotometer.

MATERIALS AND METHODS

STANDARD OPERATIVE PROCEDURE

COLLECTION AND AUTHENTICATION OF THE RAW DRUGS

The raw drug was procured from Ramasamy chetty shop, Paris, Chennai and authenticated by competent authority of Department of Gunapadam, National Institute of Siddha, Chennai.

INGREDIENTS[29]:

Purified Karkadagashigi (Rhus succedanea)
Purified Siruthekku (Clerodendrum serratum)
Purified Amanaku Ver (Ricinus communis)
Purified Kandangattari Ver (Solanum surratense)
Purified Jadamanjil (Nardostachys grandiflora)
Purified Chukku (Zingiber officinale)
Purified Milagu (Piper nigrum)
Purified Thippili (Piper longum)
Purified Kadukkai (Terminalia chebula)
Purified Thantrikkai (Terminalia bellirica)
Purified Indhuppu (Sodium chloride impura)

All are in equal proportions.

PURIFICATION PROCESS :

Purification of Karkadagashingi[30]:

The raw drug was purified by roasted in the almond oil.

Purification of Siruthekku[30]:

The raw drug was purified by removed the outer layer and cut into small pieces. Finally the pieces are dried in the sunlight.

Purification of Amanaku ver[30]:

The raw drug was washed well in water.

Purification of Kandangattari ver[30]:

The raw drug was washed well in water.

Purification of Jadamanjil[30]:

The raw drug was purified by dried in the sunlight.

Purification of Chukku[9]:

The raw drug was purified by soaked in the limestone water and the outer layer was removed.

Purification of Milagu[9]:

The raw drug was soaked in buttermilk for 1hour 15 minutes and then roasted the drug.

Purification of Thippili[9]:

The raw drug was purified by soaked in the lemon juice.

Purification of Kadukkai[9]:

The raw drug was purified by removed the seeds.

Purification of Thantrikkai[9]:

The raw drug was purified by removed the seeds.

Purification of Indhuppu[31]:

The raw drug was purified by soaked in goat's urine for 3 nazhigai (72 minutes) and dried in the sunlight.

PREPARATION OF THE MEDICINE[29]:

The raw drug was purified and pulverized by an electric grinder into fine powder, separately. And then it was sieved by using a fine silk cloth (vasthrakayam). The fine powder was mixed with milk and backed in a backing pan (pittaviyal method). Then it was dried and ultra filtered by a cotton cloth and made into fine powder again. The powder was stored in a clean, dry air tight glass container.

LABELLING:

Name of the preparation	: Singathi Chooranam
Quantity of the drug	: 14 gm
Dose	: 1 gm, bid
Adjuvant/ vehicle	: Luke warm water
Indication	: Eraippu [Bronchial Asthma]
Date of manufacturing	: The drug was prepared in two batches. 14/06/2012, 04/09/2012.
Date of expiry	: 3 months from the date of manufacture.

மிளகு

(Piper nigrum)

தான்றி

(Terminalia bellirica)

கடுக்காய்

(Terminalia chebula)

சுக்கு

(Zingiber officinale)

திப்பிலி

(Piper longum)

சடாமாஞ்சி

(Nardostachys grandiflora)

கர்க்கடகசிங்கி

(Rhus succedanea)

ஆமணக்கு

(Ricinus communis)

கண்டங்கத்திரி

(Solanum surattense)

கண்டுபாரங்கி

(Clerodendrumserratum)

இந்துப்பு

(Sodium chloride impura)

(Before purification)

(Sodium chloride impura)

(After purification)

SINGATHI CHOORANAM

REVIEW OF LITERATURE

GUNAPADAM ASPECT[8]

கர்க்கடகசிங்கி (Rhus succedanea, Linn.)

வேறுபெயர் : கர்க்கடகசிங்கி, கற்காடகசிங்கி.

சுவை : துவர்ப்பு, **தன்மை:** வெப்பம், **பிரிவு :** கார்ப்பு.

செய்கை : துவர்ப்பி, உரமாக்கி, உடலுரமாக்கி, செரிப்புண்டாக்கி, கோழையகற்றி, வெப்பமுண்டாக்கி,

குணம் :

"கர்க்கடக சிங்கி கபங்காசம் ஈளையொடு
முக்கல் கிராணி முதிரிரைச்சல் - பொக்கெனவே
காடுகின்ற பேதியையுஞ் சாடும் அரிவையரைக்
கூடுதிறங் கொடுக்குங் கூறு."

இதனால் ஐயத்தாலுண்டாகும் இருமல், சற்றுக்கூடிக் கழியும் நிணக்கழிச்சல் நோய், குருதிக் கழிச்சலிலுண்டாகும் கடுப்பு, ஈளை, வயிற்றிரைச்சல் இவைகள் நீங்கும்.

கண்டுபாரங்கி (Clerodendrum serratum (Linn) Moon)

வேறுபெயர் : சிறுதேக்கு

பயன்படும் உறுப்புகள்: வேர்

சுவை : கைப்பு, துவர்ப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு.

செய்கை : வெப்பமுண்டாக்கி, தாதுவெப்பகற்றி

குணம் :

"கண்டுபா ரங்கியெனுஞ் சிறுதேக் குண்டேல்,
காலெங்கே பித்தமெங்கே கபந்தா னெங்கே
தொண்டுதொட்டுத் தொடர்சுவாச காச மெங்கே
சுரமெங்கே வெறியெங்கே தொனிநோ யெங்கே
மிண்டுபுரி பீநசநீர்க் கோவை யெங்கே
வெளிநீருண் ணீரெங்கே விறற்கா லெங்கே
அண்டுபாடச் சீதசுரங் கடுப்பு மெங்கே
யழலையக நோயெங்கே யறைகு வீரே!"

இதனால் முக்குற்றம், இரைப்பிருமல், உள்வெப்பு, வெறி நோய் (பைத்தியம்), சுரம், மூக்கில் முன்நீர்க்கோவை, பின் நீர்க்கோவை, நாட்பட்ட வளிநோய், குளிர் காய்ச்சல், உடல்வலி, உட்காந்தல், மனத் தடுமாற்றம் தீரும்.

ஆமணக்கு (Ricinus communis)

வேறுபெயர் : ஏரண்டம், சித்திரம், தலரூபம்.

பயன்படும் உறுப்பு : வேர்

சுவை : கைப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு.

செய்கை : வாதமடக்கி

குணம் :

“வாதத் தொடக்கை வரவொட்டா மற்படிக்குக்
காதத்துக் கப்பாற் கடியுமே - சூதத்தைப்
பேரண்டப் பந்திக்கும் பேதிக்கு நோய்க்கட்டை
யேரண்ட மென்பதினியே” - தேரன் வெண்பா

பொருள் :

கழிச்சலை உண்டாக்கி வளிக்குற்றத்தை எழவொட்டாமற்றடுக்கும்.

கண்டங்கத்திரி (Solanum surattense, Burm.f.)

பயன்படும் உறுப்புகள் : வேர்

சுவை : கார்ப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு

செய்கை : கோழையகற்றி, சிறுநீர்ப்பெருக்கி, அகட்டுவாய்வகற்றி.

குணம் :

“வேரிலைபூ காய்பழமவ் வித்துமதன் பட்டையுமிவ்
வூரி லிருக்க உடற்கனப்பும் - நீராய்
வரும்பீந சங்கயஞ்சு வாசமுந்தங் காதே
அருங்கண்டங் கத்தரியு ளார்”

உடலின் நீரேற்றம், மூக்கில் நீர் பாய்தல், ஈளை (கயம்), இரைப்பு இவை போம்.

சடாமாஞ்சி (Nardostachys grandiflora, DC.)

வேறுபெயர் : சடாமாசி, ஜடமாஞ்சி, பைசாசி, சடிலை, மாமிசி, பூதசேசிநி

பயன்படும் உறுப்புகள் : வேர்

சுவை : (பச்சையில்) இனிப்பு (காய்ந்தபின்) கார்ப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு.

செய்கை :வெப்பமுண்டாக்கி, இசிவகற்றி, சிறுநீர்ப்பெருக்கி, கோழையகற்றி

குணம் :

"குட்டஞ் சிலந்திவிடம் கோர புராண சுரம்
உட்டினங்கால் பேதிகண்ணோய் ஒட்டிருமல் - சொட்டிரத்த
பித்தமிரைப் பேகும் பெருங்கோரை என்றுரைக்குஞ்
சுத்தசடா மாஞ்சிலை சொல்"

இதற்கு சிலந்தி நஞ்சு, பழையசுரம், உட்சூடு, வாய்வு, கழிச்சல், கண்ணோய், இருமல், குருதியழல், இரைப்பு நீங்கும்.

சுக்கு (Zingiber officinale, Rosc.)

வேறுபெயர் : அருக்கன், ஆர்த்ரகம், கடுபத்திரம், சுண்டி சொண்டி, நாகரம், விடமூடிய அமிர்தம்

பயன்படும் உறுப்புகள்: கிழங்கு (உலர்ந்தது)

சுவை : கார்ப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு.

செய்கை : வெப்பமுண்டாக்கி, பசித்தீத்தூண்டி, அகட்டுவாய்வகற்றி

குணம் :

"சூலைமந்தம் நெஞ்செரிப்பு தோடமேப் பம்மழலை
மூலம் இரைப்பிருமல் மூக்குநீர் - வாலகப
தோடமதி சார தொடர்வாத குன்மநீர்த்
தோடம்ஆ மம்போக்குஞ் சுக்கு"

சுக்கினால், செரியாமை, மார்பெரிச்சல், புளியேப்பம், வெப்பம், கீழ்வாய் நோய், இரைப்பு, இருமல், கழிச்சல், நீரேற்றம், குன்மம், வயிற்றுப்பிசம், காதுக் குத்தல், முகநோய், தலை நோய், குலைவலி, பாண்டு, வயிற்றுக் குத்தல், ஐயசுரம் போம்.

மிளகு (Piper nigrum. Linn)

வேறுபெயர் : கலினை, கறி,காயம், திரங்கல், மிரியல், மாசம், மலையாளி

பயன்படும் உறுப்புகள்: பழம்

சுவை : கைப்பு, கார்ப்பு, **தன்மை :** வெப்பம், **பிரிவு :** கார்ப்பு.

செய்கை : காறலுண்டாக்கி, அகட்டுவாய்வகற்றி, முறைவெப்பகற்றி, வெப்ப முண்டாக்கி, நச்சரி

குணம் :

"சீதசுரம் பாண்டு சிலேத்மங் கிராணிகுன்மம்
வாதம் அருசிபித்தம் மாமூலம் - ஓதுசன்னி
யாசமபஸ் மாரம் அடன்மேகம் காசமிவை
நாசங் கறிமிளகினால்."

குளிர்சுரம், பாண்டு, கோழை, குன்மம், வாயு, சுவையின்மை, வெறி, மூலம், இருமல், செவிவலி, செரியாமை இவை போகும்.

திப்பிலி (Piper longum)

வேறுபெயர் : காமன், குடாரி, கோழையகற்றி, கனை, கலினி, வைதேகி, அம்பு

பயன்படும் உறுப்புகள் : காய், அரிசி

சுவை : இனிப்பு, **தன்மை** - தட்பம், **பிரிவு** - இனிப்பு.

உலர்ந்தது - கார்ப்பு, தன்மை - வெப்பம், பிரிவு - இனிப்பு.

செய்கை :வெப்பமுண்டாக்கி, அகட்டுவாயுவகற்றி

குணம் :

"ஈளை யிரும லிரைப்புப் பசப்பிணிகள்
மாள வொழியாமல் வாட்டுமே - யாளுமுறை
பாங்கா யறிந்துசெய்வீர் பண்டிதத்தைப் பண்டிதரே
வேங்கைவாய்ப் பான்கணை மெய்" (தேரன் வெண்பா)

ஈளை, இருமல், இரைப்பு, உப்பிசம் முதலிய பிணிகளைப் போக்கத் தக்கவாறு திப்பிலியை ஆளவேண்டியதாம்.

கடுக்காய் (Terminalia chebula)

வேறுபெயர் : அமுதம், வனதுர்க்கி, ரோகிணி, ஜீவநிகா, ஜீவந்தி, அரிதகி, அம்மை.

பயன்படும் உறுப்பு : தோல்

சுவை : முக்கிய சுவை - துவர்ப்பு, அத்துடன் சிறிது - இனிப்பு, புளிப்பு, கார்ப்பு, கைப்பு

தன்மை :வெப்பம் **பிரிவு :** இனிப்பு

குணம் :

"தாடை கழுத்தக்கி தாலு குறியிவிடப்
பீடை சிலிபதமுற் பேதிமுடம் - ஆடையெட்டாத்
தூலமிடி புண்வாத சோணிகா மாலையிரண்
டாலமிடி போம்வரிக்கா யால்."

கடுக்காயினால் கன்னம், கழுத்து, நா, ஆண்குறி, இவ்விடங்களில் நோய்கள், காலடிப்புற்றுநோய், அதிதூலம், இடிப்புண், வாதசோணிதம், காமாலை, தாவர, சங்கமவிடங்கள் இவை போம்.

தான்றி (Terminalia bellirica (Gaertn.) Roxb.)

வேறுபெயர் :

அம்பலத்தி, எரிகட்பலம், கந்தகட்பலம், கூலித்துருமம், களந்தூன்றி, பூதவாசகம்

பயன்படும் உறுப்புகள்: இலை, பழம், விரை

சுவை : துவர்ப்பு **தன்மை :** வெப்பம் **பிரிவு :** இனிப்பு

செய்கை :துவர்ப்பி, கோழையகற்றி, மலமிளக்கி, உரமாக்கி.

குணம் :

"சிலந்திவிடம் காமியப்புண் சீழான மேகங்
கலந்துவரும் வாதபித்தங் காலோ - டலர்ந்துடலில்
ஊன்றிக்காய் வெப்ப முதிரபிந் துங்கரக்குந்
தான்றிக்காய் கையிலெடுத் தால்."

இதனால் சிலந்திநஞ்சு, ஆண்குறிப்புண், வெள்ளை, குருதியழல் நோய், வளி தீ குற்றங்களால் வரும் நோய்கள் போம்.

இந்துப்பு (Sodium chloride impura)[31]

வேறுபெயர்கள் :

சைந்தவம், சிந்தூரம், மதிகூர்மை, மதியுப்பு, மிந்தாச்சொல், சந்திரனுப்பு

சுத்தி :

இதனைக் காடியில் மூன்று நாள் ஊறப்போட்டு, சூரியனொளியில் உலர்த்தி எடுக்கச் சுத்தியாகும்.

இதனைக் காடி அல்லது வெள்ளாட்டு நீரில் மூன்ற நாழிகை மத்தித்து வெய்யிலில் உலர்த்திக் கொள்ள இது சுத்தியாகும்.

செய்கை :

மலகாரி, பசித்தீத்தூண்டி, அகட்டுவாய்வகற்றி, சிறுநீர்ப்பெருக்கி

குணம் :

“சென்னிக்கண்ணா பற்றூர் செவிகவுள்கண் டம்கபநோய்
சந்நியா சங்காசந் தாகமிரைப் - புன்னிரத்த
மூலஞ் சிலந்திநளி மூடிகளுஞ் சூதை வலி
சூலஞ் சிதையுமிந்தாற் சொல்”

அட்டகுன்மம், மந்தம், அசிர்க்கரம், கபபித்தம், மலக்கட்டு, காமிய நோய், கரப்பான், ஐயநோய், நேத்திரகாசம், தாகம், சுவாசம், இரத்த மூலம் முதலிய பிணிகள் நீங்கும்.

சேரும் மருந்துகள்

கற்கடகசிங்கி சேரும் மருந்துகள்

திரிபலாதி சூரணம்[10]:

அளவு : திரிகடிப் பிரமாணம் அனுபானம் : தேன்

தீரும் நோய்கள் : சீதள சம்பந்தமான ஈளை, இருமல், காசம்

கண்டகாரிக் கிருதம்[10]:

அளவு : 2 - 3 தேக்கரண்டி

தீரும் நோய்கள் : உளைமந்தை, சுவாசகாசம், ஈளை, இருமல், சயம்

சிறுதேக்கு சேரும் மருந்துகள்

திப்பிலி இலேகியம்[10]:

அளவு : சுண்டைக்காய்ப் பிரமாணம்

தீரும் நோய்கள் : ஈளை, இருமல், காசம், சயம், வாந்தி

ஆடாதோடைச் சூரணம்[30]:

ஆளவு : 1/4 தோலா, அனுபானம் : பால்

தீரும் நோய்கள் : சீதள சம்பந்தத்தாலுண்டான சுவாசகாசம்

தாளிசாதி சூரணம்[32]:

அளவு : 1 - 2 கிராம், அனுபானம் : தேன்

தீரும்நோய்கள் : கபநோய் - 96, இருமல், தொண்டைக் கட்டு

கண்டங்கத்திரி வேர் சேரும் மருந்துகள்

மகா சிஞ்சாதி இலேகியம்[29]:

அளவு : 1 - 2 தேக்கரண்டி

தீரும்நோய்கள் : குன்மம், பாண்டு, சோபை, சுவாசகாசம்

சூத்திர அபையாதி லேகியம்[29]:

அனுபானம் : கடுக்காய் குடிநீர்

தீரும்நோய்கள் : சூலை, சுவாசகாசம், பாண்டு, குன்மம்

மகர சுதர்சணச் சூரணம்[30]:

அளவு : 1/2 தோலா, அனுபானம் : வெந்நீர்

தீரும்நோய்கள் : கபசுரம், சுவாசகாசம், காமாலை

சடாமாஞ்சில் சேரும் மருந்துகள்

மூதண்ட லேகியம்[10]:

அளவு : கழற்சிக் கொட்டை பிரமாணம்

தீரும் நோய்கள் : சுவாசம், சஷயம், காசம், நீர்க்கோவை, சூலை

இலவங்காதி சூரணம்[30]:

அளவு : 1/2 தோலா

தீரும் நோய்கள் : இரைப்பு, மயக்கம், சுரம், இருமல்

தூதுளைக் கிருதம்[10]:

அளவு : 1 - 2 தேக்கரண்டி

தீரும் நோய்கள் : ஈளை, இருமல், சுவாசம், இரத்த காசம்

சுக்கு சேரும் மருந்துகள்

சத்திக்சாரணைக் கிருதம்[30]:

அளவு : 1/2 - 1 கரண்டி

தீரும் நோய்கள் : காசம், சுவாசகாசம், சுரம்

சவுபாக்கிய சுண்டி[29]:

அளவு : சுண்டையளவு

தீரும் நோய்கள் : சயம், அக்கினிமந்தம், காசம், சுவாசம்

திராட்சாதிச் சூரணம்[32]:

அளவு : 1 - 2 கிராம், அனுபானம் : தேன்

தீரும் நோய்கள் : இருமல், சுவாசகாசம், பாண்டு

மிளகு சேரும் மருந்துகள்

சுவாச குடோரி சூரணம்[10]:

அளவு : திரிகடிப் பிராமாணம், அனுபானம் : தேன், இஞ்சிச்சாறு

தீரும்நோய்கள் : ஈளை, இருமல், கோழை வாந்தி

வில்வாதி இலேகியம்[10]:

அளவு : பாக்களவு

தீரும் நோய்கள் : குன்மம், காசம், ஈளை, இருமல்

காச குலாந்தக மாத்திரை[10] :

அனுபானம் : முலைப்பால், இஞ்சிச்சாறு

தீரும்நோய்கள் : சுவாசம், காசம், ஈளை

திப்பிலி சேரும் மருந்துகள்

இராம மகேசுரம் குளிகை[33]:

அளவு : குன்றியெடை

தீரும்நோய்கள் : சயம், ஈளை, குன்மம், ஐயம்

திப்பிலி சூரணம்[10]:

அளவு : திரிகடிப் பிரமாணம், அனுபானம் : தேன்

தீரும்நோய்கள்: இருமல், ஈளை, சுவாசகாசம்

பஞ்சகோல கிருதம்[10]:

அளவு : 2 - 3 தேக்கரண்டி, அனுபானம் : கற்கண்டு பொடி

தீரும்நோய்கள் : ஈளை, இருமல், செரியாமை

கடுக்காய் சேரும் மருந்துகள்

மஹா மதனகாமேசுர இலேகியம்[30]:

அளவு : கழற்சிக்காய் பிரமாணம்

தீரும்நோய்கள் : ஈளை, இருமல், சூலை

மேக சிந்தாமணி மெழுகு[30]:

அளவு : சுண்டைக்காயளவு

தீரும்நோய்கள் : குன்மம், சுவாசகாசம், இசிவு

தாபவனலச் சூரணம்[29]:

அனுபானம் : பசு நெய்

தீரும் நோய்கள் : சன்னி, சுவாசகாசம், சயம், இருமல்

தான்றிக்காய் சேரும் மருந்துகள்

கல்யாண கிருதம்[30]:

அளவு : 1 கரண்டி

தீரும்நோய்கள் : சுரம், சஷயம், சுவாசகாசம்

வல்லாரை நெய்[32]:

அளவு : 1 - 2 தேக்கரண்டி

தீரும்நோய்கள் : ஈளை, இரைப்பு, சுவாசகாசம்

இந்துப்பு சேரும் மருந்துகள்

ஆடாதோடைக் கிருதம்[10]:

அளவு :1 தேக்கரண்டி

தீரும்நோய்கள் : நெஞ்சுவலி, கபச்சிக்கலால் உண்டான இரைப்பு, சுவாசகாசம்

ஆறுமுகச் செந்தூரம்[10]:

அளவு : 1/2 - 1 குன்றியெடை அனுபானம் : தேன்

தீரும்நோய்கள் : சுரம், ஈளை, சஷயம், காசம்

மகா அக்கினி குமாரன்[29]:

அளவு : 1 - 2

தீரும்நோய்கள் : சுவாசகாசம், 13 வகை சன்னி, வாத குன்மம், சூலை

BOTANICAL ASPECTS[34]

கர்க்கடகசிங்கி

BOTANICAL NAME : Rhus succedanea Linn

VERNACULAR NAME

ENG	: Galls
SANS	: Karkatashring
HINDI	: Kakarsingi
TEL	: Karkatakashrungi
MAL	: Karkatasringi

CLASSIFICATION[12]

Kingdom	: Plantae
Class	: Dicotyledons
Subclass	: Polypetalae
Series	: Disciflorae
Order	: Sapindales
Family	: Anacardiaceae
Subfamily	: Anacardioideae
Genus	: Rhus (syn. Searsia)
Species	: R.succedanea

BOTANICAL DESCRIPTION : Leaves alternate, pinnate, 15-23 cm long, with or without terminal leaflet 4-5 pairs, lanceolate, acuminate, sub-opposite, coriaceous, 7-12 cm long. Characteristic Galls are produced on leafy branches. Flowering: March-May; Fruiting: June-October.

PARTS USED : Leaf galls

PHYSICAL CONSTANTS : Foreign matter – Not more than 2%; Total ash- Not more than 7%; Acid- insoluble ash- Not more than 0.2%; Alcohol- soluble extractive- Not less than 30%; Water- soluble extractive- Not less than 30%.

CHEMICAL CONSTITUENTS : Resin, pistacienoic acids A and B, tannins, β-sitosterol, α-piene, β-piene,dl-limonene, dihydroqercetin.

PHARMACOLOGICAL ACTIVITIES : Expectorant, antispasmodic, antibacterial, antiinflammaory, antiallergic, antimicrobial, antifungal.

SUPPORTIVE JOURNAL ARTICLES[35]:

Rhus Succedanea shown significant effect on constrictor response of histamine, acetylcholine and serotonin on smooth muscles.

கண்டுபாரங்கி

BOTANICAL NAME : Clerodendrum serratum(Linn)

VERNACULAR NAME

ENG	: Beetle Killer
SANS	: Bharngi, Barbara
HINDI	: Bharangi
TEL	: Gantu sharag
MAL	: Cerutekku

CLASSIFICATION[12]

Kingdom	:Plantae
Class	:Dicotyledons
Subclass	:Gamopetalae
Series	:Bicarpellatae
Order	:Lamiales
Family	:Verbenaceae
Genus	:Clerodendrum
Species	:C.serrata

BOTANICAL DESCRIPTION : Perennial herbs or shrubs, 0.9-2.4 m high. Leaves sessile or nearly so, opposite or sometimes ternate, passing upwards into bracts, narrowly obovate-oblong or sub-elliptic, acute or acuminate, usually coarsely and sharply serrate. Flowers many, blue-purple or white, arranged in dichotomous cymes, the whole forming

a lax, subpyramidal panicle. Drupes 6 mm long, broadly obovoid, rather succulent, dark-purple when ripe.

PARTS USED : Root

CHEMICAL CONSTITUENTS : Serratagenic acid, queretaroic acid, Ferulic acid, scutellarein baicalein.

PHARMACOLOGICAL ACTIVITIES: Antihistaminic, bronchoconstrictor, antiallergic, antiasthmatic, antibiotic, stomachic.

SUPPORTIVE JOURNAL ARTICLES[36] :

Icosahydropicenic acid (IHPA), a new pentacyclic triterpenoid saponin was first time isolated from the roots of Clerodendrum Serratum (L) Moon (Verbenaceae). IHPA, at the dose of 100mg/kg, showed significant protection of mast cell degraualtion (59.62%) as compared to standard sodium cromoglycate (64.48%). The compound also revealed significant inhibitory activity on histamine—induced goat tracheal chain preparation. The study provides scientific basis for its clinical use in the treatment of asthma.

ஆமணக்கு

BOTANICAL NAME : Ricinus communis

VERNACULAR NAME

ENG : Castor, Castor oil plant
SANS : Yeranda
HINDI : Erandi
TEL : Erandamu
MAL : Avanakku

CLASSIFICATION[12]

Kingdom : Plantae
Class : Dicotyledons
Series : Unisexuals
Order : Malpighiales
Family : Euphorbiaceae

Subfamily : Acalyphoideae

Genus : Ricinus

Species : R.communis

BOTANICAL DESCRIPTION : Tall annuals, sometimes shrubby or tree-like. Leaves alternate, broad, palmately 5-11-lobed, serrate. Flowers monecious, in terminal subpaniculate racemes, 30-60 cm long. Fruit a pickly capsule of three 2-valved cocci. Seeds oblong, testa crustaceous, variously coloured, mottled.

PARTS USED : Root

PHYSICAL CONSTANTS : **Root –** Ash – Not more than 8.0%; Acid insoluble ash – Not more than 1.0%; Alcohol soluble extractive – Not less than 3.0%; Water soluble extractive – Not less than 9.0%.

CHEMICAL CONSTITUENTS : Germanicol ester derivative and an unidentified triterpene; Inorganic material like potassium, sodium, magnesium, chloride, nitrate, iron, aluminium, manganese, calcium, carbonate and phosphate including gallotannins.

PHARMACOLOGICAL ACTIVITIES : Anti-inflammatory, spasmogenic, purgative, immunizing and spasmolytic.

SUPPORTIVE JOURNAL ARTICLES[37] :

The Methanolic Extract of Ricinus Communis root at a dose of 250 mg/kg p.o exhibited significant Anti inflammatory activity in carrageenin induced rat paw edema model and a higher dose of 500 mg/kg p.o also exhibited significant activity in cotton pellet granuloma model in Wister Albino rats.

கண்டங்கத்திரி

BOTANICAL NAME : Solanum surattense, Solanum virginianum

VERNACULAR NAME

ENG : Yellow-berried nightshade

SANS : Kanta-karika Nidegdhika

HINDI : Kateli

TEL : Nelamulaka, Vakudu

MAL : Kantankattiri

CLASSIFICATION[12]

Kingdom	: Plantae
Class	: Dicotyledons
Subclass	: Gamopetalae
Series	: Polemoniales
Order	: Solanales
Family	: Solanaceae
Genus	: Solanum
Species	: S.xanthocarpum

BOTANICAL DESCRIPTION : Procumbent or trailling herb or under shrub with many branches clothed with 10-15 mm long prickles. Leaves ovate or elliptic, sinuate or sub-pinnatifid, prickly. Flowers blue or violet-purple, in lateral cymes. Berries globose, 1.2-2 cm in diameter, glabrous, yellow or whitish with green markings. Seeds glabrous, subreniform, 2-2.5 mm in diameter.

PARTS USED : Whole plant

PHYSICAL CONSTANTS : Total ash – 2.881%; Acid soluble ash- 0.645%; Water soluble extractive- 15.32%; Ethanol soluble extractive- 12.27%; Pet ether soluble extractive- 5.33%.

CHEMICAL CONSTITUENTS : Carpesterol, gluco-alkaloid solanocarpine, solasodine, solasonine, solamargine, coumarins, scopolin, scopoletin, esculin and esculetin.

PHARMACOLOGICAL ACTIVITIES : Antibiotic, antiinflammatory, analgesic, antifungal, diuretic, spasmolytic, antipyretic.

SUPPORTIVE JOURNAL ARTICLES[38] **:**

Apigenin has shown anti allergic effect in ovalbumin induced asthma.

Ova induced mice showed allergic airway reactions and included an increase in number of eosinophils in bronchoalveolar lavage fluid, an increase in inflammatory cell infiltration into lung around blood vessels and airways, airway luminal narrowing and development of airway hyper responsiveness.

Administration of Apigenin before last airway ova challenge resulted in a significant inhibtion of all asthmatic reactions.

சடாமாஞ்சி

BOTANICAL NAME : Nardostachys grandiflora DC, Nardostachys jatamansi DC

VERNACULAR NAME

ENG	: Valerina root
SANS	: Jatmansi
HINDI	: Jatamansi
TEL	: Jatamamsi
MAL	: Manchi

CLASSIFICATION[12]

Kingdom	: Plantae
Order	: Dipsacales
Family	: Valerianaceae
Genus	: Nardostachys
Species	: N.grandiflora

BOTANICAL DESCRIPTION : An erect perennial, aromatic herb, 10-70 cm high, with long, stout, woody greyish, rhizomatous, tail-like rootstock covered with reddish-brown hairs or tufted fibrous remains of the petioles of withered radical leaves. Radical leaves 15-20 x 2.5 cm, longitudinally nerved, glabrous, narrowed into the petiole; cauline leaves 1 or 2 pairs, 2.5-7.5 cm, sessile, oblong or subovate. Flowers pale-white or pink, rosy in terminal corymbose cymes.

PARTS USED : Rhizome

CHEMICAL CONSTITUENTS : Actinidine, carotene,nardol, nardostachonol, isovaleric acid, valeranone, β-sitosterol, jatamansone, jatmansic acid, oroselol, oroselone.

PHARMACOLOGICAL ACTIVITIES : Hypotensive, analgesic, tranquillising, antibacterial, antifungal, diuretic, antimicrobial, antiulcerogenic, antianxiety, bronchodilatory.

SUPPORTIVE JOURNAL ARTICLES[35] :

The alcoholic extract of rhizomes of Jatamansi exhibited bronchodialatory effects in guinea pig, induced by histamine.

The extract also showed antispasmodic effect in experimental models on the isolated smooth muscles.

Extract of Nardostachys jatamansi shown significant effect on constrictor response of histamine, acetylcholine and serotonin on smooth muscles.

சுக்கு

BOTANICAL NAME : Zingiber officinale Rose

VERNACULAR NAME

ENG	: Dried Ginger
SANS	: Nagaram
HINDI	: Sonth
TEL	: Sonthi
MAL	: Chukku

CLASSIFICATION[12]

Kingdom	: Plantae
Division	: Magnoliophyta
Class	: Monocotyledon
Order	: Zingiberales
Family	: Zingiberaceae
Genus	: Zingiber
Species	: Z.officinale

BOTANICAL DESCRIPTION : Perenial herb; rhizome stout, tuberous with erect leafy stem, 10-25 x 1.5-3 cm, narrowed to the base, acute or acuminate; sheath 10-15 cm long.

PARTS USED : Dried rhizome

PHYSICAL CONSTANTS : Dried rhizome – Total ash – Not more than 6%; Water soluble ash – Not less than 1.5%; Alcohol (90%) soluble extractive – Not less than 3%; Water soluble extractive- Not less than 10%.

CHEMICAL CONSTITUENTS : β- sesquiphellandrene, gingerol, zingerone, shogaol, diarylheptenones, c, two diarylhepatonoids, Camphene, phellandrene, cineol, citral, borneol, zingiberene, gingerol, shogaol and gingediol.

PHARMACOLOGICAL ACTIVITIES : Antiinflammatory, hypolipidaemic, antiemetic, antiulcer, antiplatelet, antipyretic, antioxidant, antibacterial, antifungal, hypoglycaemic.

SUPPORTIVE JOURNAL ARTICLES[39] :

Ginger suppresses prostaglandin synthesis through inhibition of cyclooxygenase-1 and cyclooxygenase-2. Ginger also suppresses leukotriene biosynthesis by inhibiting 5-lipoxygenase.

This pharmacological property distinguishes ginger from nonsteroidal anti-inflammatory drugs.

மிளகு

BOTANICAL NAME : Piper nigrum Linn

VERNACULAR NAME

ENG	: Black pepper, common pepper
SANS	: Maricha
HINDI	: Kalimirch
TEL	: Miriyalu
MAL	: Kurumulaku

CLASSIFICATION[12]

Kingdom	: Plantae
Division	: Magnoliophyta
Class	: Dicotyledons
Subclass	: Magnoliidae
Order	: Piperales
Family	: Piperaceae

Genus : Piper

Species : P.nigrum

BOTANICAL DESCRIPTION : Fruits ovoid or globose, one seeded, bright red when ripe. Seeds globose, testa thin, perisperm hard and white.

PARTS USED : Fruit

PHYSICAL CONSTANTS : Total ash- Not more than 5.0%; Acid insoluble ash- Not more than 0.5%; Alcohol soluble extractive- Not less than 6.0%; Water soluble extractive- Not less than 6.0%.

CHEMICAL CONSTITUENTS : Piperonal, piperine, piperoleine A&B, α-pinene, sabiene, β-pinene, myrcene, p-cymene, pellitonine, piperettine, piperonal,alamine, arginine.

PHARMACOLOGICAL ACTIVITIES: Antioxidant, sedative, analgesic, muscle relaxant, antipyretic, antiinflammatory, antifungal, antimicrobial, antiulcer, antibacterial.

SUPPORTIVE JOURNAL ARTICLES[40] :

To evaluate the anti-asthmatic activity of Piper nigrum on acetylcholine induced contraction of goat tracheal chain preparation. The aqueous extract of Piper nigrum fruits at the doses of 380 mcg/ml and 640 mcg/ml significantly inhibited acetylcholine induced bronchoconstriction of isolated goat trachea. Thus the present study revealed that the aqueous extract of fruit of Piper nigrum has significant anti-asthmatic potential.

திப்பிலி

BOTANICAL NAME : Piper longum Linn

VERNACULAR NAME

ENG : Indian long pepper, Long pepper

SANS : Pippali

HINDI : Pipli

TEL : Pipallu

MAL : Tippali

CLASSIFICATION[12]

Kingdom : Plantae
Division : Magnoliophyta
Class : Dicotyledons
Order : Piperales
Family : Piperaceae
Genus : Piper
Species : P.longum

BOTANICAL DESCRIPTION : Fruits ovoid, yellowish orange, sunk in fleshy spike.

PARTS USED : Fruit

CHEMICAL CONSTITUENTS : Piperlongumine and piperlonguminine; Piperine and sesamin; Sesquiterpene hydrocarbon, caryophyllene, piperine, pipernonaline and piperundecalidine; Sylvatin, sesamin and diaeudesmin.

PHARMACOLOGICAL ACTIVITIES : Antibacterial, antiinflammatory, antimalarial, antitubercular, insecticidal, antispasmodic, cough-suppressor, immunostimulatory, anthelmintic, antinarcotic, antiulcerogenic.

SUPPORTIVE JOURNAL ARTICLES[41] **:**

The extracts of the fruits of piper longum (100 μg $Ml^{-1)}$ significantly ($p<0.01$) inhibited the histamine induced contraction of isolated Guinea pig ileum preparation. The extracts (50, 100, 200 mg kg^{-1}) showed the significant ($p<0.01$) activity and increase in dose of extract increased the % protection in histamine induced bronchospasm and also showed significant ($p<0.01$) activity in haloperidol induced catalepsy and passive paw anaphylaxis.

கடுக்காய்

BOTANICAL NAME : Terminalia chebula Retz

VERNACULAR NAME

ENG : Chebulik myrobalan
SANS : Pathya, Haritaki

HINDI : Hara

TEL : Karakkaya

MAL : Katukka

CLASSIFICATION[12]

Kingdom : Plantae

Division : Magnoliophyta

Class : Magnoliopsida

Order : Myrtales

Family : Combretaceae

Genus : Terminalia

Species : T.chebula

BOTANICAL DESCRIPTION : Drupes ellipsoidal, obovoid or ovoid, yellow to orange-brown, sometimes tinged with red or black and hard when ripe, 3-5 cm long, 5 ribbed on drying. Seeds hard, pale yellow.

PARTS USED : Fruit

PHYSICAL CONSTANTS : Foreign matter – Not more than 1%; Total ash – Not more than 5%; Acid insoluble ash – Not more than 5%; Alcohol soluble extractive – Not less than 40%; Water soluble extractive – Not less than 60%.

CHEMICAL CONSTITUENTS : Anthraquinone glycoside, chebulinic acid, chebulagic acid, tannic acid, terchebin, tetrachebulin, vitamin C, arachidic, behenic, linolec, oleic, palmitic and stearic acids.

PHARMACOLOGICAL ACTIVITIES : Antimicrobial, antifungal, antibacterial, antistress, sntispasmodic.

SUPORTIVE JOURNAL ARTICLES[42]**:**

Terminalia chebula has been reported to exhibit, Anti tussie property, Anti histamine property.

The water and ethanolic extracts of the fruit on Guinea pig ileum, were found to have strong anti histaminic activity.

தான்றி

BOTANICAL NAME : Terminalia bellirica(Gaertn.) Roxb

VERNACULAR NAME

ENG : Belliric Myrobalan

SANS : Vebeethaki

HINDI : Bhaira

TEL : Tandra

MAL : Tanni, Tannikka

CLASSIFICATION[12]

Kingdom : Plantae

Division : Magnoliophyta

Class : Magnoliopsida

Order : Myrtales

Family : Combretaceae

Genus : Terminalia

Species : T.bellirica

BOTANICAL DESCRIPTION : A large tree, upto 40 m high. Fruits globular, 1.5 – 2.5 cm in diam, obscurely 5- angled when dry.

PARTS USED : Fruit

PHYSICAL CONSTANTS : Foreign matter – Not more than 2%; Total ash – Not more than 7%; Acid insoluble ash – Not more than 1%; Alcohol soluble extactive – Not less than 8%; Water soluble extractivev – Not less than 35%.

CHEMICAL CONSTITUENTS : Chebulagic acid, ellagic acid, mannitol and rhamnose, β- sitosterol and bellericanin, palmitic, oleic and linoleic acids.

PHARMACOLOGICAL ACTIVITIES : Purgative, antifungal, antihistaminic, antiasthmatic, broncho-dilatory, anti-spasmodic antibacterial, antistress.

SUPPORTIVE JOURNAL ARTICLES[43] **:**

The extract of terminalia bellirica fruit was found to possess antihistaminic effect on experimental asthma in Guinea –pigs. The anti asthmatic effect was confirmed by pollen-induced asthma in animals.

MINERALOGICAL ASPECT

இந்துப்பு (ROCK SALT)

Halite, commonly known as Rock salt, is the mineral form of Sodium chloride[44].

VERNACULAR NAMES[45]:

ENG : Rock salt, Bay salt

SANS : Saindhava

HINDI : Sendhalon

TEL : Saindhalavanam

MAL : Intu-uppu

GUJ : Sindhaluna

OCCURENCE:

It occurs mostly in United states of America, Canada, New mexico, Islamabad, Pakistan, United kingdom[44].

This is taken out from earth especially in the North West regions of Punjab and Sind(Pakistan)[46].

CHARACTERS[44]:

It is colourless or white but may also be light blue, purple, pink, red, orange, yellow or grey depending on the amount and the type of impurities. It is saline in taste. It commonly occurs with minerals such as sulfates, halides and borates.

PHYSIOCHEMICAL PROPERTIES[44]:

Category	- halide mineral
Chemical formula	- NaCl
Crystal symmetry	- Isometric hexoctahedral
Molar mass	- 58.433 g/mol
Colour	- colourless or white
Crystal system	- Cubic
Luster	- Vitreous
Streak	- White
Optical properties	- Isotropic
Refractive index	- 1.544
Solubility	- water soluble

PURIFICATION[46]:

Rock salt was kept soaked in Vinegar (old rice fomented water) for three days and insolated to get purified and detoxified form.

The rock salt was kept soaked in Goat's urine for three days and insolated to get purified form.

ACTIONS[45]**:**

Carminative

Stomachic

Digestive

Cathartic

Emetic.

USES[45]**:**

It is given in dyspepsia and other abdominal disorders.

Apart from the medicinal uses it is used for managing ice and in the cooking purposes.

PHYSICAL PROPERTIES

Materials and Methods

The Physical properties of Singathi Chooranam were analysed in the following procedure. It was done at Sri Ramachandra University, Chennai.

pH at 10% of aqueous solution:

Five grams of Singathi Chooranam was weighed accurately and placed in clear 100 ml beaker. Then 50 ml of distilled water was added to it and dissolved well. After 30 minutes it was then applied in to pH meter at standard buffer solution of 4.0,7.0,9.2. (Trial drug II, Table 2)

Ash Values

The Ash values measures of the inorganic constituents present in the raw drug. A high ash content explains its unsuitable nature to be used as a drug. (Trial drug II,Table 2)

Total Ash

A little of extract was taken in a silica crucible previously ignited, cooled and weighed. It was incinerated by gradually increasing the heat not exceeding dull red heat (450°C) until free from carbon, cooled and weighed. The percentage of ash was calculated with reference to air- dried drug. The procedure was repeated to get the constant weight. (Trial drug II, Table 2)

Water soluble ash

The total ash was boiled with 25 ml water and filtered through ash less filter paper (Whatmann 4.1). It was followed by washing with hot water .The filter paper was dried and ignited in the silica crucible, cooled and the water insoluble ash was weighed. The water-soluble ash can be calculated by subtracting the water insoluble ash from the total ash. (Trial drug II, Table 2)

Acid insoluble ash

The total ash obtained was boiled for 5 minutes with 25 ml of (10% w/v) dilute hydrochloric acid and filtering through ash less filter paper (Whatmann 4.1). The filter paper was ignited in the silica crucible, cooled and insoluble ash was weighed. (Trial drug II, Table 2)

HIGH PERFORMANCE THIN LAYER CHROMATOGRAPHY

The HPTLC of Singathi Chooranam was done at Sri Ramachandra University, Chennai.

HPTLC Fingerprint - RH1

SAMPLE PREPARATION

100 mg of extract was weighed and dissolved in 70% methanol to get a concentration of 10mg/ml concentration this is then used for injection.

CHROMATOGRAPHIC CONDITION FOR HPTLC FINGER PRINT (Trial drug II, Graph 1)

SampleName	:	Singathi chooranam
Sample-ID	:	105
Stationary phase	:	Silica gel F 254
Mobile phase	:	n-Hexane: Ethyl acetate: Formic acid 60:40:2.5 ml)
Scanning wavelength	:	254,298,489 nm
Sample concentration	:	20 mg/ml
Injecting volume	:	5, 10 µl
Development mode	:	Ascending mode

Significance of HPTLC fingerprinting in Standardisation

Standardisation of traditional medicine has become mandatory in the present national and international scientific scenario, as they have to stand competing with stringent regulatory methods and also clinically. HPTLC is one of the versatile chromatographic methods presently available for the rapid analysis of herbal drugs due to several reasons. Firstly the time required for the demonstration of the most of the characteristic constituents of a drug is very quick and short. Secondly, in addition to qualitative detection, HPTLC also provides semi-quantitative information on the major active constituents of a drug, thus enabling an assessment of drug quality. Thirdly the fingerprint obtained is suitable for monitoring the identity and purity of drugs and for detecting adulteration and substitution. Hence in order to check the identity, purity and standardise the quantity of active principles in the herbal extracts a HPTLC fingerprint of all the ingredient medicinal plants used in the formulation has been obtained.

The distribution of phyto-constituents in a plant depends on various factors such as soil, time of collection period of storage, etc. So, it is necessary to standardize the extract being used for pharmacological studies. HPTLC serves as a convenient tool for finding out the distribution pattern of phyto constituents which is unique to each plant. The HPTLC finger-printing profile establishes the identity and purity of the raw drug being used. It helps in the authentification of the plant material.

Chromatographic Conditions

The finger printing has been done using the following chromatographic conditions. Chromatography was performed on a10x10 cm pre activated HPTLC silica gel 60F 254 plate. Samples were applied to the plate as 6mm wide band with an automatic TLC applicator Linomat 5 with N2 flow (CAMAG, Switzerland), 8mm from the bottom. Densitometric scanning was performed on CAMAG scanner III.The plates were pre-washed by methanol and activated at 60^0 C for 5 minutes prior to chromatography. The slit dimension was kept at 5 minutes x 0.45 minutes and 20 minutes scanning speed was employed. The mobile phase was chosen after running each plant in different mobile phases of varying polarity (Toluene, Toluene: Ethyl acetate and Ethyl acetate: Methanol) and 10 ml of mobile phase was used per chromatography. Linear ascending development was carried out in 20 cm x 10-em twin glass chamber saturated with the mobile phase.

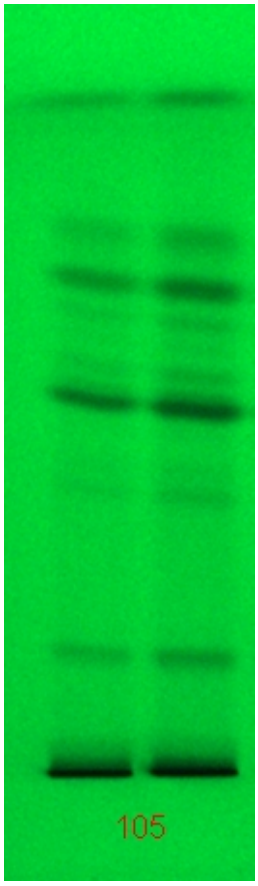
Chromatographic Analysis

The hydro alcoholic extracts of the plants have been prepared at a concentration of 10 mg/10 ml in alcohol and were spotted using CAMAG Linomat 5 applicator. The method was optimized by selecting appropriate mobile phase for respective plant extracts and developed in a twin trough chamber, 20 x 10 cm at 25°C. The plates were dried by hair dryer. The developed plates were scanned at appropriate wavelength using CAMAG TLC scanner 3 and photo-documented using CAMAG REPROSTAR 3.

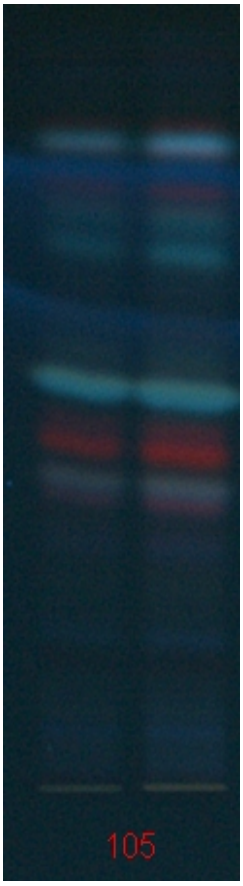
Inference

HPTLC fingerprint of RH -1 shows four peaks at Rf values 0.25, 0.31, 0.41 & 0.95. The peak correspond to the Rf value 0.31 has maximum peak area of 7256.5. At this stage it is difficult to confirm the individual components present in the extract, but

from our lab experience on phytochemical analysis, we suggest that the major peaks found in the fingerprint may be acidic glycosides / resins. Since, in the present chromatographic conditions, the above mentioned components will be eluted easy.



254nm (*No: 105 -13*)



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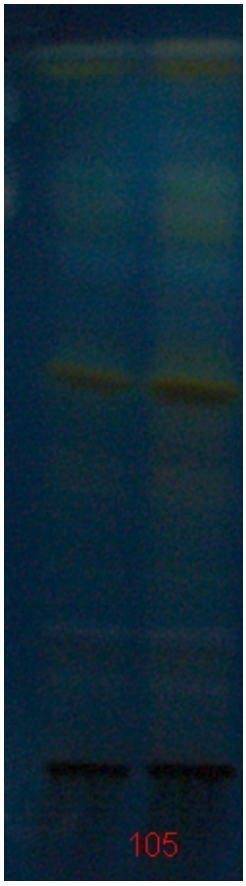
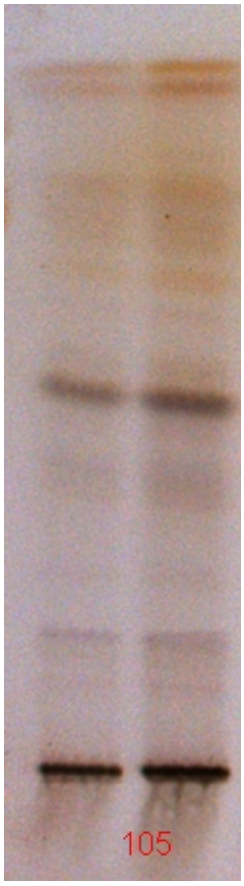


366nm (*No: 105 -15*)



White light (*No: 105 -16*)

BIO -CHEMICAL ANALYSIS OF SINGATHI CHOORANAM

The biochemical analysis of the Singathi Chooranam was carried out in the Biochemistry lab, National Institute of Siddha, Chennai.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
1.	Appearance of sample	Light yellow in colour	
2.	**Solubility:** a. A little(500mg) of the sample was shaken well with distilled water. b. A little(500mg) of the sample was shaken well with con. HCl/Con. H_2So_4	Sparingly soluble	Absence of Silicate
3.	**Action of Heat:** A small amount(500mg) of the sample was taken in a dry test tube and heated gartly at first and then strong.	No white fumes evolved	Absence of Carbonate
4.	**Flame Test:** A small amount(500mg) of the sample was made into a paste with con. HCl in a watch glass and introduced into non-luminous part of the Bunsen flame.	No Bluish green flame appeared.	Absence of Copper
5.	**Ash Test:** A filter paper was soaked into a mixture of sample and dil. cobalt nitrate solution and introduced into the Bunsen flame and ignited.	Yellow colour flame appeared.	Presence of sodium

Preparation of Extract:

5gm of Singathi Chooranam was weighed accurately and placed in a 250ml clean beaker and added with 50ml of distilled water. Then it was boiled well for about 10 minutes. Then it was cooled and filtered in a 100ml volumetric flask and made up to 100ml with distilled water.

S.No	EXPERIMENT	OBSERVATION	INFERENCE
	I.Test For Acid Radicals		
1.	**Test For Sulphate:** a. 2ml of the above prepared extract was taken in a test tube to this added 2ml of 4% dil ammonium oxalate solution b. 2ml of the above prepared extracts was added with 2ml of dil-HCl was added until the effervescence ceases off. Then 2ml of dil.Barium chloride solution was added.	Cloudy appearance present	Absence of Sulphate
2.	**Test For Chloride:** 2ml of the above prepared extract was added with dil. HCl till the effervescence ceases. Then 2ml of dil.silver nitrate solution was added.	No cloudy appearance.	Presence of Chloride
3.	**Test For Phosphate:** 2ml of the extract was treated with 2ml of dil.ammonium molybdate solution and 2ml of con.HNo3.	Yellow appearance present	Absence of Phosphate

4 .	**Test For Carbonate:** 2ml of the extract was treated with 2ml dil. Magnesium sulphate solution	No Cloudy appearance.	Absence of carbonate
5.	**Test For Nitrate:** 1gm of the substance was heated with copper turning and concentrated H2So4 and viewed the test tube vertically down.	No Brown gas evolved.	Absence of Nitrate
6.	**Test For Sulphide:** 1gm of the substance was treated with 2ml of con. HCL	No Rotten Egg Smelling gas.	Absence of Sulphide
7.	**Test For Fluoride & Oxalate:** 2ml of extract was added with 2ml of dil. Acetic acid and 2ml dil.calcium chloride solution and heated.	No Cloudy appearance	Absence of fluoride and oxalate
8.	**Test For Nitrite:** 3drops of the extract was placed on a filter paper, on that-2 drops of dil.acetic acid and 2 drops of dil.Benzidine solution was placed.	No Characteristic changes	Absence of Nitrite
9.	**Test For Borate:** 2 Pinches(50mg) of the substance was made into paste by using dil.sulphuric acid and alcohol (95%) and introduced into the blue flame.	No Bluish green colour flame.	Absence of borate

	II. Test For Basic Radicals		
1.	**Test For Lead:** 2ml of the extract was added with 2ml of dil.potassium iodine solution.	No yellow Precipitate obtained.	Absence of Lead
2.	**Test For Copper:** a. One pinch(50mg) of substance was made into paste with con. HClin a watch glass and introduced into the non-luminuous part of the flame.	No Blue colour flame No Blue colour precipitate formed.	Absence of copper
3.	**Test For Aluminium:** To the 2ml of extract, dil.sodium hydroxide was added in 5 drops to excess.	NoYellow colour appeared.	Absence of aluminium
4.	**Test For Iron:** a. To the 2ml of extract,2ml of dil.ammonium solution was added. b. To the 2ml of extract 2ml thiocyanate solution and 2ml of con HNo3 was added	blood red colour appeared.	Presence of Iron
5.	**Test For Zinc:** To 2ml of the extract, dil.sodium hydroxide solution was added in 5 drops to excess and dil.ammonium chloride was added.	No White precipitate was formed	Absence of Zinc
6.	**Test For Calcium:** 2ml of the extract was added with 2ml of 4% dil.ammonium oxalate solution	No Cloudy appearance and white precipitate was obtained	Absence of calcium
7.	**Test For Magnesium:** To 2ml of extract dil.sodium hydroxide solution was added in drops to excess.	White precipitate was obtained	Presence of Magnesium

8.	**Test For Ammonium:** To 2ml of extract 1 ml of Nessler's reagent and excess of dil.sodium hydroxide solution are added.	No Brown colour appeared	Absence of ammonium
9.	**Test For Potassium:** A pinch(25mg) of substance was treated of with 2ml of dil.sodium nitrite solution and then treated with 2ml of dil.cobalt nitrate in 30% dil.glacial acetic acid.	No Yellowish precipitate was obtained.	Absence of Potassium
10.	**Test For Sodium**: 2 pinches(50mg) of the substance was made into paste by using HCl and introduced into the blue flame of Bunsen burner.	yellow colour flame appeared	Presence of sodium
11.	**Test For Mercury**: 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No yellow precipitate was obtained	Absence of mercury
12.	**Test For Arsenic:** 2ml of the extract was treated with 2ml of dil.sodium hydroxide solution.	No brownwish red precipitate was obtained	Absence of arsenic

	III. Miscellaneous		
1.	**Test For Starch:** 2ml of extract was treated with weak dil.iodine solution	No blue colour developed	Absence of starch
2.	**Test For Reducing Sugar:** 5ml of Benedict's qualitative solution was taken in a test tube and allowed to boil for 2 minutes and added 8 to 10 drops of the extract and again boil it for 2 minutes. The colour changes are noted.	Brick red colour not developed	Absence of reducing sugar
3.	**Test For The Alkaloids:** a) 2ml of the extract was treated with 2ml of dil.potassium iodide solution. b) 2ml of the extract was treated with 2ml of dil.picric acid. c) 2ml of the extract was treated with 2ml of dil.phosphotungstic acid.	. Yellow colour developed	- Presence of Alkaloid
4.	**Test For Tannic Acid:** 2ml of extract was treated with 2ml of dil.ferric chloride solution	No black precipitate was obtained	Presence of Tannic acid
5.	**Test For Unsaturated Compound:** To the 2ml of extract 2ml of dil.Potassium permanganate solution was added.	Potassium permanganate was not decolourised	Absence of unsaturated compound

6.	**Test For Amino Acid:** 2 drops of the extract was placed on a filter paper and dried well. 20ml of Biurette reagent was added.	violet colour developed	Presence of amino acids
7.	**Test For Type Of Compound:** 2ml of the extract was treated with 2 ml of dil.ferric chloride solution.	No green colour developed No red colour developed No violet colour developed No blue colour developed	Absence of oxy quinole pinephrine and pyro catechol Anti pyrine, Aliphatic amino acids and meconic acid are absent Apomorphine salicylate and Resorcinol are absent Morphine, Phenol cresol and hydro uinone are absent

ELEMENTAL ANALYSIS USING ATOMIC ABSORPTION SPECTROPHOTOMETER

The AAS of Singathi Chooranam was done at Sri Ramachandra University, Chennai.

Elemental analysis using atomic absorption spectrophotometer

In this method the sample, in the form of a homogeneous liquid, is introduced into a flame where thermal and chemical reactions create "free" atoms capable of absorbing, emitting or fluorescing at characteristic wavelengths.

In Atomic Absorption Spectrophotometer (AAS) the majority of free atoms in the commonly used flames were in the ground state, but that the flames did not also have enough energy to excite these atoms. A light source emitting a narrow spectral line of the characteristic energy is used to excite the free atoms formed in the flame. The decrease in energy (absorption) is then measured.

METHODOLOGY

I. Microwave Digestion For Elemental Analysis

Model Name: Multiwave3000

Digestion Procedure:

200mg of the given sample is placed in a digestion vessel, acid is added and the mixture is heated for several minutes. After the digestion, the samples are diluted to a specific volume. If too much sample is used in wet digestion, the reaction mixture can become violent. The samples are placed in digestion vessels that fit directly into digestion racks. There are several different acids or mixtures of acids used for digestion, the most common of which is concentrated Hydrochloric acid. The samples are heated slowly at a high temperature. After digestion, the samples are diluted to the appropriate volume with deionized H_2O.

II. Elemental Analysis using Atomic Absorption Spectrophotometer

The elemental analysis of digested samples have been determined by Atomic Absorption Spectrophotometer- Flame technique (AAS model 400 Perkin Elmer). Working standard solutions of Fe were prepared from stock standard solution of 1000 ppm from MERCK. Using blank solution to zero the instrument performs the Calibration. The standards are then analyzed and their absorbance recorded. A graph of Absorbance Vs Concentration is plotted. Calibration of the instrument was repeated periodically during operation. A blank reading was also taken and necessary correction was made during the calculation of concentration of various elements.

The digested material was made upto 100 ml for analysis in an (AAS) atomic absorption spectrophotometer (Perkin Elmer). The results were calibrated using standard calibration curve.

In AAS the wave Length(nm), Flame type, Lamp source and Calibration range (ppm) of different elements have been used, are listed in table.

Instrumental conditions for elemental analysis

Element	Wavelength nm	Light source	Flame type
Iron	386.0	HCL	Air/Ac

Air/Ac: Air-Acetylene; HCL: Hallow cathode lamp

Elemental Analysis using Flame photometer

The analysis of Na of the digested samples have been determined by Flame photometer (Flame photometer 129- Systronics Make). (Trail drug II, Table 4)

TOXICITY STUDY

ACUTE AND SUB ACUTE TOXICITY STUDY OF SINGATHI CHOORANAM IN RODENTS

Animals

Mice of either sex weighing 25-30g and rats weighing 210-240g were obtained from the animal house of Vels University. The animals were used with the approval of the Institute Animal Ethics Committee and obtained from Vels University, Chennai. They were fed with a balanced standard pellet diet and maintained under standard laboratory conditions, providing 24-28^{0}C temperature, standard light cycle (12 h light, 12 h dark) and water ad libitum.

Animals were kept in cages with raised floors of wide mesh to prevent coprophagy. Animal welfare guidelines were observed during the maintenance period and experimentation. The rats were randomly assigned to control and different treatment groups, six animals per group. The animals were acclimatized for one week under laboratory conditions.

ACUTE TOXICITY STUDY-OECD 425 GUIDELINES

Acute oral toxicity test for the Singathi Chooranam was carried out as per OECD Guidelines 425. As with other sequential test designs, care was taken to ensure that animals are available in the appropriate size and age range for the entire study. The test substance is administered in a single dose by gavage using a stomach tube or a suitable intubation cannula. The fasted body weight of each animal is determined and the dose is calculated according to the body weight. After the substance has been administered, food was withheld for a further 2 hours in mice. The animals were observed continuously for the first 4 h and then each hour for the next 24 h and at 6 hourly intervals for the following 48 h after administering of the test drug, to observe any death or changes in general behaviour and other physiological activities.

Single animals are dosed in sequence usually at 48 h intervals. However, the time interval between dosing is determined by the onset, duration, and severity of toxic signs.

Treatment of an animal at the next dose was delayed until one is confident of survival of the previously dosed animal.

Observation of toxicity signs: General behavior, respiratory pattern, cardiovascular signs, motor activities, reflexes, change in skin and fur, mortality and the body weight changes were monitored daily. The time of onset, intensity, and duration of these signs, if any, was recorded.

SUB-ACUTE TOXICITY

In a 28-days sub acute toxicity study, twenty four rats of either sex were divided into four groups of 6 rats each. Group I that served as normal control was administered with distilled water (p.o.) while groups II, III and IV were administered daily with the Singathi Chooranam (p.o.) for 28 days at a dose of 100, 200 and 400 g/kg respectively. The animals were then observed daily for gross behavioural changes and any other signs of subacute toxicity.

The weight of each rat was recorded on day 0 and weekly throughout the course of the study, food and water consumption per rat was calculated. At the end of the 28 days they were fasted overnight, each animal was anaesthetized with diethylether, following which they were then dissected and blood samples were obtained by cardiac puncture into heparinised tubes. The blood sample collected from each rat was centrifuged with 3000 X g at $4^{o}C$ for 10 min to separate the serum and used for the biochemical assays.

Hematological and blood biochemical analysis:

At the end of the study, all animals were kept fasted for 16-18 h and then anesthetized with anesthetic ether on the 28th day. Blood samples for hematological and blood chemical analyses were taken from retro orbital vein. Heparinized blood samples were taken for determining complete blood count (white blood cell count, differential white blood cell count, platelet count, red blood cell count, hematocrit, and hemoglobin) by semiautomated hematology analyzer.

The serum from non-heparinized blood was carefully collected for blood chemistry and enzyme analysis like glucose, Creatinine, Total protein, Albumin, Total

and Direct bilirubins, Serum glutamate-oxaloacetate transaminase (SGOT), Serum glutamate pyruvate transaminase (SGPT), and Alkaline phosphatase (ALP) were automatically determined using autoanalyzer.

Necropsy:

All rats were sacrificed after the blood collection. The positions, shapes, sizes and colors of internal organs were evaluated. The Spleen, Testes, Pancrea, Lung, Liver, Brain, Heart, Stomach, Intestine, Bone, Ovary, and Kidney tissues were excised from all rats to visually detect gross lesions, and weighed to determine relative organs' weights and preserved in 10% neutral formalin for histopathological assessment. The tissues were embedded in paraffin, and then sectioned, stained with haematoxylin and eosin and were examined microscopically.

Statistical analysis

Values were represented as mean ± SEM. Data were analysed using one-way analysis of variance (ANOVA) and group means were compared using the Tukey-Kramer Multiple Comparison test using Instat-V3 software. P values < 0.05 were considered significant.(Table 5 – Table 12)

RESULTS

Animals were not shown any significant toxic clinical signs during the dosing period of 28 days. All animals from control and all the treated dose groups survived throughout the dosing period of 28 days and it was found one animal dead after 24days of treatment in high dose. Results of body weight determination of animals of control and different dose groups exhibited comparable body weight gain throughout the dosing period of 28 days.

During dosing period, the quantity of food consumed by animals from different dose groups was found to be comparable and normal with that of control animals. Ophthalmoscopic examination of animals in control and Singathi Chooranam treated groups did not reveal any major and remarkable abnormality. These tests conducted on the experimental animals at termination and recorded did not reveal any abnormalities.

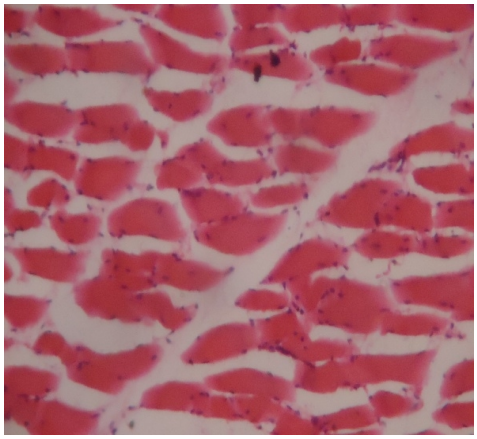
Urine analysis data of control group and treated group of animals determined did not reveal any abnormalities.

Comparison of organ weights of treated animals with respective control animals on day 28 was found to be comparable. Gross pathological examination of animals in control as well as the treated groups did not reveal any abnormalities. The results of haematological investigations conducted on day 28, revealed, the increase or decrease in the values obtained was within normal biological and laboratory limits. Results of Biochemical investigations revealed the following significant changes in the values of different parameters were within normal biological and laboratory limits.

CONCLUSION

From the results obtained in this study, no toxic effect was observed upto 400mg/kg of Singathi Chooranam via oral route over a period of 28 days. So, it can be concluded that the Singathi Chooranam can be prescribed for therapeutic use in human with the dosage recommendations of upto 400mg/kg. body weight p.o.

HISTOPATHOLOGICAL SLIDES



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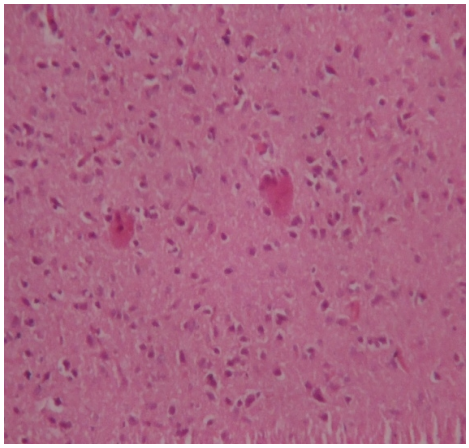

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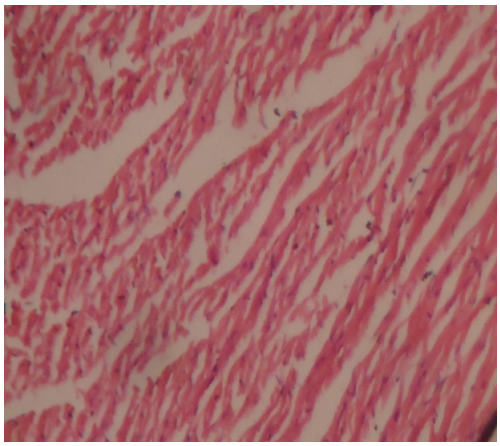

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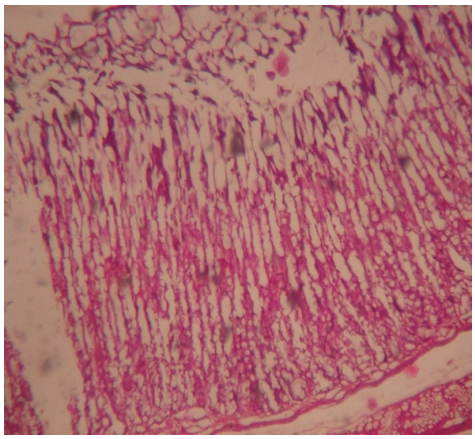

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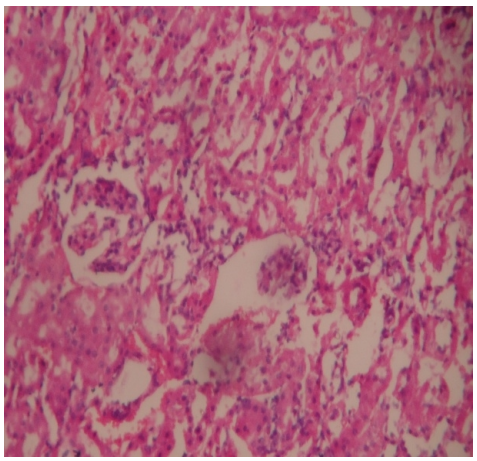
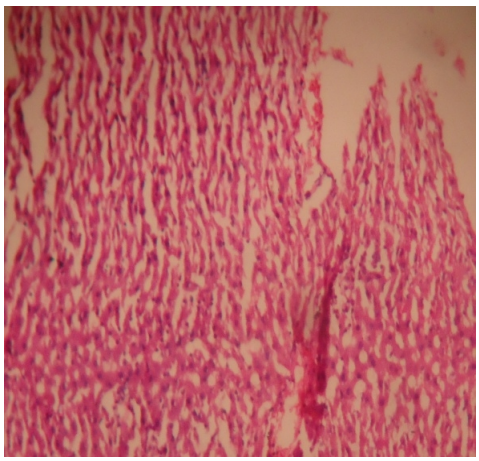
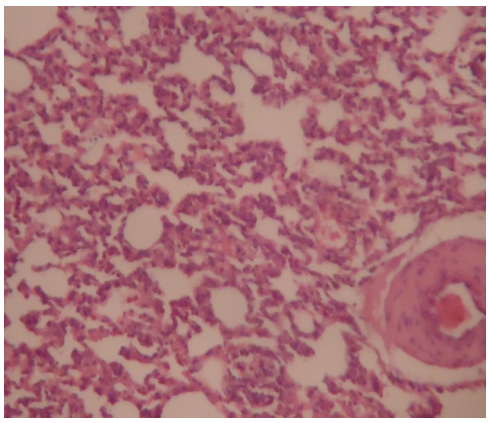


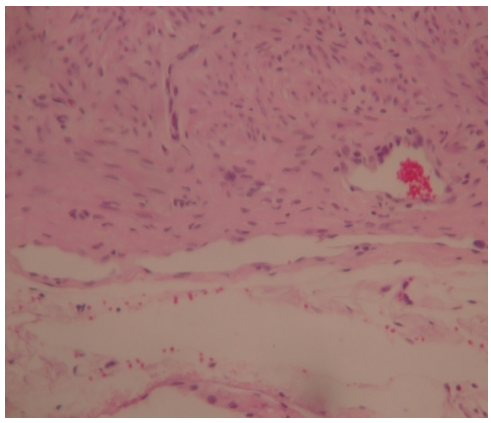
KIDNEY



LIVER



LUNG



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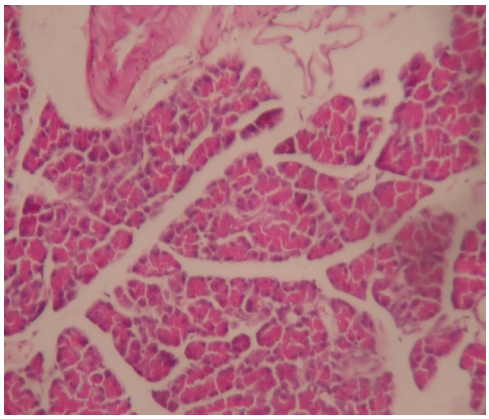

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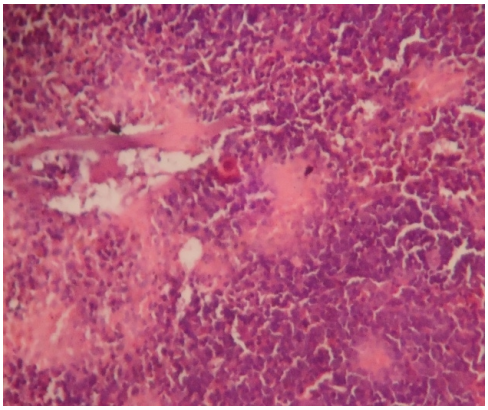

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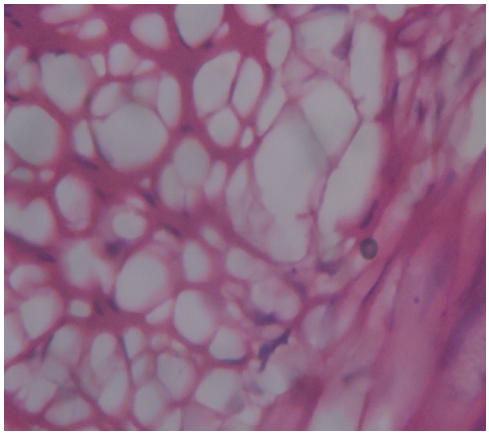
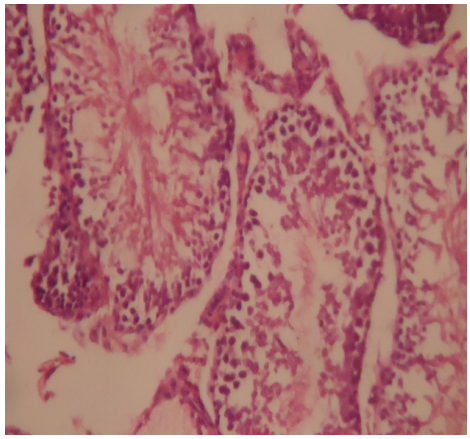


STOMACH



TESTIS

PHARMACOLOGICAL STUDIES

MATERIALS AND METHODS

Drugs And Stock Solution

Drugs used were Histamine diphosphate (Sigma Chemical, USA) and Promethazine hydrochloride (Rhone – Poulenc, Mumbai). Histamine dihydrochloride was dissolved in distilled water and desired concentrations were prepared. The test drug Singathi chooranam concentration was 100 microgram per ml prepared by suspending with 2% CMC and then the volume was adjusted to 10 ml with normal saline for making the concentration of 100 μg/ml.in distilled water.

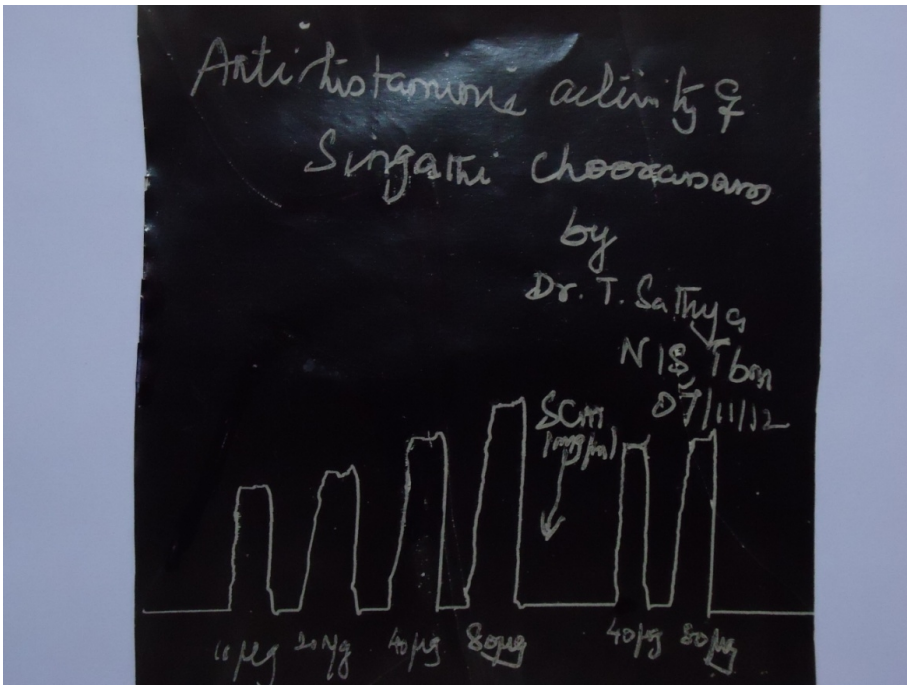
Animals

Male albino guinea pig weighing 350– 400g was kept in fasting condition 18 hours prior to commencement of experiment and given water ad libitum. It was housed under standard laboratory conditions of temperature (25 ± 2°C) and 12/12 hr light/dark cycle and then sacrificed by a blow to the head and exsanguinated as per CPCSEA recommended guidelines. (XIII/VELS/PCOL/39/2000/CPCSEA/IAEC/08.08.2012)

IN-VITRO ANTIHISTAMINIC STUDY

Guinea pig was sacrificed and a segment from ileum (2 cm) was dissected from the terminal ileum and mounted in an organ bath containing Tyrode solution (10 ml) between two stainless steel hooks under 0.5 to 1 g initial tension. The lower hook was fixed at the bottom of the organ bath and upper one was connected to an isotonic transducer. The Tyrode solution composition (pH 7.4) was (concentration in gm/lit.) NaCl 8.0, KCl0.2, CaCl2 0.2, MgCl2 0.1, NaHCO3 1.0, NaH2PO4 0.05, and Glucose1.0 gm/liter. It was continuously aerated and maintained at 37 ± 0.5ºC The equilibrium period was 60 min and the bath solution was refreshed every 15 min. After equilibrium period, a dose response curve for histamine in variant molar concentrations, by maintaining 45 min time cycle.

ANTIHISTAMINIC ACTIVITY



BRONCHODILATOR STUDY

Animals were divided into four groups of six animals each. Each animal were served as its own control. Animals belonging to each group were subjected to a histamine aerosol (0.2% Histamine diphosphate in saline) using a glass nebuliser for 2 sec in an airtight Perspex chamber. Aerosolization of the solution was achieved via a compressed air line operating at a pressure of 8 Psi and a flow rate of 5ml/min. After exposure to the histamine aerosol, the animal showed signs of immediate immobilization and bouts of coughing. This was followed by shallow breathing symptoms, after which the animal collapsed, fell on its back and convulsed. The time taken by the animal to fall on its back after exposure to the aerosol was designated as the exposition time. The exposition time for each animal in all the four groups was noted.

Once the animal fell on its back, it was immediately taken out of the chamber and exposed to fresh air where the animal returned back to normal. After 1 hour the animals in the first three groups were administered orally 100, 200 and 400 mg/kg p.o, of Singathi chooranam respectively. While the fourth group of animals received 300µg/kg of

Promethazine by oral route. One hour later, the animals were reexposed to the aerosol and exposition time for each animal was noted. The difference in the exposition time before and after Singathi chooranam administration was taken as a measure of the protective effect. Percent protection afforded by the Singathi chooranam was calculated by the formula.

$$\text{Percentage Protection} = \frac{\text{Eta-Etb}}{\text{Etb}} \times 100$$

Where 'Eta' is the mean exposition time after treatment with extract and 'Etb' is the mean exposition time before treatment with extract.

STATISTICAL ANALYSIS

Ileum contractions induced by agonist were assumed as 100% and reductions induced by test drug calculated. Percentage of ileum contraction was expressed as mean ± SEM. Results were analyzed using one-way analysis of variance (ANOVA). Probability value less than 0.05 were considered as significant. (Trial drug II, Table 13 & 14, Graph 2 & 3)

RESULTS AND DISCUSSION

Bronchial asthma is characterized by wide spread narrowing of the bronchial tree due to contraction of smooth muscle and releases series of chemical mediators including histamine, leukotrienes etc. Histamine when inhaled has been shown to induce bronchoconstriction by direct H1 receptor activation and also by a neutrally mediated bronchoconstrictor effect via vagal reflexes. The mean increase in exposition time against histamine challenge was significantly ($P<0.01$) increased with increasing doses of Singathi chooranam and offered dose dependent protection 20.21% (200mg/kg) and 26.06% (400mg/kg). Whereas known anti histaminic drug Promethazine hydrochloride (300µg/kg, p.o) also inhibited histamine induced bronchoconstriction in guinea pigs and offered 40.75% of protection against histamine challenge. Histamine has shown to activate action potentials in the intrapulmonary vagal afferents. The inflammatory response is characterized by an increase in the numbers of eosinophils and mast cells, mucus hypersecretion, and activation of T cells. Several studies have shown that T-helper type (Th2) cells play a major role in the initiation and maintenance of allergic airway

inflammation and asthma through their increased production of Th2-type cytokines (IL-4, IL-5, and IL- 13). These inflammatory cytokines also produced in the bronchial tissue by mast cells, alveolar macrophages, and epithelial cells, play a significant role in the pathogenesis of airway inflammation. Histamine contracts the guinea pig ileum muscle of guinea pig. Guinea pig ileum is easier to handle and to prepare; it is also much more sensitive.

Therefore, the dose relative contractile responses of different agonists like ACh, histamine, 5hydroxytryptamine and bradykinin can be observed in isolated Guinea pig ileum. In the present study the isolated guinea pig ileum preparation; there is right side shift of Dose Response Curve of histamine in the presence of Singathi chooranam indicating Bronchodilation. Thus, the present study provides the experimental evidence for the presence of bronchodilator effect of Singathi Chooranam.

CONCLUSION

Based on the results of the above study, it can be concluded that the Singathi chooranam possesses significant antihistaminic and bronchodilating activity.

DISEASE ASPECT

SIDDHA ASPECT

இரைப்பு நோய்[13]

வேறுபெயர்கள் : இழுப்பு நோய், சுவாசகாசம், சுவாசம், ஈளை. சுரம்

இயல்பு :

இந்நோய் இன்ன வகைத்தெனக் குறிப்பிடக் கூடாதபடி, ஒரு காரணமுமின்றி மார்பை வலித்து இறுக்கியது போன்று வேதனையைத் தந்து, மூச்சை வெளிவிடவும், உள் இழுக்கவும் முடியாமல் திணறச் செய்யும். அன்றியும், வெளியாகும் மூச்சு மிகுந்த சிரமத்தோடு வெளியாவதுடன், குழல், யாழ், வீணை முதலிய வாத்தியங்களின் ஒலியைப் போல் ஒலிக்கும். மேலும், இருமலால் மார்பிலுள்ள சளியை வெளியாக்குவதற்கு இயற்கை முயலினும் அக்கோழை வெளியாதல் இல்லை.

நோய் வரும் வழி :

இயற்கையாயமைந்த உடலினுக்கு, வேண்டாத உணவு, செயல் முதலியவைகளால், வன்மை குறைந்த நிலையில் ஐயத்தை மிகுதிபடுத்தக் கூடிய உணவாலும், நடத்தையாலும், புல், பூண்டு, அரிசி, கேழ்வரகு, முதலியவைகளின் சுணையாலும், தனக்கு உதவாத நாற்றப் பொருள்களை முகர்வதாலும் இந்நோய் பிறக்கும்.

நோயின் முற்குறிகள் :

தங்களுக்கு ஆகா உணவும், ஆகா காற்றின் மணமும் பட்டவுடன் மூக்கில் நீர்பாய்தல், தும்மல் வருதல், மாப்பு நோதல், மார்பை இறுக்கிக் கட்டியது போல உண்டாதல், இயற்கை மூச்சுத் தட்டுபடல், விலாப்பக்கம் வலித்து மூச்சுத் திணறல்.

வகைகள் :

வளி இரைப்பு நோய் :

மூச்சு விடுவதற்கு வேண்டிய உடல் வன்மை இல்லாது, விடும் மூச்சு தன்வலிவு குறைந்து எழும். நெஞ்சினுள் யாதும் இல்லாதது போன்ற ஓர் உணர்ச்சி தோன்றும்.

ஐய இரைப்பு நோய் :

மார்பை அடைப்பது போல் தோன்றி, மூச்சு வெளிவிட முடியாமல் மிக்க வேதனையைப் பிறப்பித்து வாட்டும். இழுப்புள்ள வேளையில் சிறிது இருமல் வாராமலும் சளியும் வெளியாகமலிருப்பின், நோயாளனை மூச்சு வெளிவிட முடியாமல் தவிக்கக் செய்து படுக்கவும் இருக்கவும் செய்யாது.

ஐயவளி இரைப்பு நோய் :

மூச்சை சரிவர உள் வாங்குவதற்கும் வெளிவிடுவதற்கும் இயலாமல் விட்டுவிட்டு மூச்செறிதலும் மலமும் நீரும் கட்டி வயிறு உப்பி பொருமுதலும் உண்டாகி உடலை வாட்டும்.

முக்குற்ற இரைப்பு நோய் :

விடும் மூச்சு பெரிய மாடு விடும் மூச்சை யொத்திருக்கும். மார்பை இறுக்கிக் கட்டியது போல நோதல், அடிக்கடி மயக்கம் உண்டாதல்.

மேல்நோக்கு இரைப்பு நோய் :

மேற்கூறிய இரைப்பு நோய்களில் எவையேனும், மருத்துவத்திற்குக் கட்டுப்படாமல், நீண்டநாள் தொடர்ந்து மேல் நோக்குக்கால் (உதானவாயு) வன்மை இழந்து மூச்சை வெளியாக்க இயலாத போது, பிணியாளன் மூச்சுத்திணறி, கண் வெளியே முட்டி, வாய் உலர்ந்து பேசமுடியாது திகைத்து படுக்கை நிலையாமல் எழுந்து வாய் திறந்தது போல மேல் நோக்கி, சற்று காற்றை வெளியாக்கி உள்ளிழுக்க அலைவான். அச்சமயத்தில் உடனே தகுந்த மருத்துவம் செய்யின் குணமடைந்து பிழைப்பான். இன்றேல், திறந்தவாய் திறந்தபடியே இருக்க, முகங்கறுத்து மயக்குமுற்று மரணத்தை அடைவான்.

குற்றம் முதலிய வேறுபாடுகள் :

ஐயக்குற்றம் தன்னிலை மாறுதலாலும், வளி அல்லது தீக்குற்றத்தையேனும் அல்லது அவ்விரு குற்றத்தையேனும் தனக்குத் துணைக்கொண்டு குறிகுணங்களைப் பிறப்பிக்கும். உடல் நிலைகளில் (ஆதாரங்களில்) மார்பிடமான அனாகதத்தில் எழும், மேல் நோக்குங்கால் (உதானன்) தன் அளவில் மிகுந்து நிற்கும்.

நாடி நடை :

“தானமுள்ள சேத்து மந்தானிளகிய

.....

..... சுவாசம் .....

பாங்கான வாதத்தில் சேத்தும நாடி

பரிசித்தால்

.....

.....

ஓங்காணும் ..... சுவாசகாசம் .... (சதக நாடி)

உற்றிடு மய்ய நாடி யோங்கியே துடித்து நின்றாற்

பற்றிடும் இரும லீளை பதறியே யிளைப்புண் டாக்கி (குணவாகடம்)

எச்சில் (கோழை சளி) :

இது நுரைத்து அளவில் மிகுந்து பளுவற்றுக் காணின், வளி (வாத)க் குற்றத்தால் வந்ததாகும்.

கறுத்து, கடினப்பட்டு புலால் மணத்துடன் காணின், ஐயக் (கபம்) குற்றத்தைக் குறிக்கும்.

வெளுத்துச் சீழ் கலந்தது போலும் மஞ்சள் நிறத்தோடும் காணின், அழல் (பித்தம்) குற்றத்தைக் காட்டும்.

MODERN ASPECT

BRONCHIAL ASTHMA[15]

Asthma is defined as a chronic inflammatory disease of airways that is characterized by increased responsiveness of the tracheobronchial tree to a multiplicity of stimuli. It is manifested physiologically by a widespread narrowing of the air passages, which may be relieved spontaneously or as a result of therapy, and clinically by paroxysms of dyspnoea, cough, and wheezing.

TYPES:

Extrinsic Asthma (Atopic asthma, Early onset asthma)

Intrinsic asthma (Non-atopic asthma, Late onset asthma)

Other types:

- Nocturnal asthma
- Gastric asthma
- Exercise induced asthma
- Episodic asthma
- Chronic asthma
- Acute severe asthma (Status Asthmatics)

Extrinsic asthma:

Onset is in childhood.

It occurs in atopic individuals who readily form IgE antibodies in response to allergens.

Intrinsic asthma:

It can begin at any age, especially in late adulthood.

There is no role for allergens in the production of the disease.

FACTORS PRECIPITATING ASTHMA:

- Cold air
- Tobacco smoke
- Dust, acrid fumes
- Emotional stress
- Respiratory infections (viral, bacterial)
- Exercise
- Drugs
  - NSAIDS especially aspirin
  - β – blockers
- Chemicals
  - Sulfiting agents like Na or K bisulfite, sulphur dioxide, etc.
- Allergens
  - Ingested (fish, nuts, strawberries)
  - Inhaled (dust, pollen, house dust mite)
  - Food additives (tartrazine, metabisulfite preservatives, ajinomoto)
- Occupational allergens
  - Grain-dust
  - Wood-dust

CLINICAL FEATURES:

- Dyspnoea
- Wheeze
- Chest pain
- Cough with mild expectoration
- Cough
- Chest tightness

COMPLICATIONS[16]:

- Status Asthmaticus
- Secondary Infection – Bronchitis, Tuberculosis
- Emphysema of lungs
- Right heart failure in late stages called chronic pulmonale.
- Bronchiectasis
- Pneumothorax

CLINICAL STUDY

The study was conducted on patients with Eraippu [Bronchial asthma] patients satisfying the inclusion criteria.

The study was conducted at the OPD/IPD of Ayothidoss Pandithar Hospital of the National Institute of Siddha, Tambaram sanatorium, Chennai-47.

Sample size:

The sample size was 20 patients.

SUBJECT SELECTION:

Inclusion criteria:

Age : 15-65 yrs

Sex : both male and female

Weight : 35-85 kgs

Patient having symptoms of

- Wheezing
- Dyspnoea
- Cough
- Cough with mild expectoration
- Chest tightness

Any of the 4 clinical symptoms

Patient who are willing to provide blood for lab investigation.

Patients who are willing to attend OPD once in 7 days.

Patient who are willing to be admitted in the hospital for 30 days.

Patient willing to sign the informed consent stating that he/she will conscientiously

stick to the treatment during 30 days but can opt out of the trial of his/her own conscious discretion.

Exclusion criteria:

Bronchiectasis

Pleural effusion

Pneumonia

Tuberculosis

Other type of asthma

Any other serious illness

Withdrawal criteria:

Development of any adverse reaction

Occurence of any other serious illness

Non co-operation of the patient

TRIAL DRUG AND DURATION

Drug : Singathi chooranam -1gm, bid with luke warm water, after food.

Duration of the treatment: 30 days.

CONDUCT OF THE STUDY:

Eraippu patients satisfying inclusion and exclusion criteria were admitted to the trial. Informed consent was obtained from the patients. Routine investigations like Blood test, urine test, and PEFR were carried out before and after the trial treatment. For in patients the drug was administered daily. For out patients the trial drug was issued for seven days course. They were advised to visit the OPD once in 7 days. During each visit they were clinically assessed.

CLINICAL OBSERVATION:

For the clinical study of "Singathi Chooranam" on Eraippu, 20 patients were selected.

Among 20 patients,11(55 %) were female, 9(45%) were male.

According to age wise distribution 40%were in 20-30 years, 30% were in 31-40 years and 30% were in 41-60 years.

Among 20 patients, All patients were affected from Wheezing, 20patients were affected from Dyspnoea, 15 patients were affected from Cough, 9 patients were affected from Expectoration and 17 patients were affected from Chest tightness.

From the clinical study 85% of patients relieved from wheezing, 70% of patients relieved from dyspnoea, 60% of patients relieved from cough, 66.67% of patients relieved from expectoration and 88.23% of patients relieved from chest tightness and no adverse effects were observed during trial period**.**

16 (80%) patients had a significant improvement in the PEFR levels after the treatment.

The following investigations were done before and at the end of the treatment.

- Blood sample (Hb, TC, DC, ESR, Sugar(F,PP), AEC, T.cholesterol,RFT, LFT)
- Peak Expiratory Flow Rate
- Urine test (Albumin, Sugar,Deposits)

The following investigations were done before treatment.

- X-ray chest - PA view
- ECG
- Sputum AFB

DISCUSSION

The drug Singathi Chooranam was selected to evaluate the Anti-shistaminic and bronchodilator activity in the treatment of Eraippu.

The literary evidence from Agathiyar Mani ennum Vaidhiya Chinthamani Venba 4000–part 1 strongly support the antihistsminic, bronchodilator activity of the drug.

Bio-chemical analysis:

The biochemical analysis of the drug reveals the presence of **sodium, magnesium, iron, chloride, alkaloids, tannic acid and amino acid.**

Magnesium[47]:

Magnesium is essential in muscle relaxation after contraction. Magnesium also plays a key role in the production of energy which is needed by the chest wall muscles and the diaphragm to perform the work of breathing.

Magnesium promotes healthy lung function by acting as a bronchodilator, preventing the bronchial passages from going into spasm. Magnesium deficiency may increase vulnerabilty to allergies by increasing the release of histamine into the bloodstream, increasing allergic reactivity in general.

Low dietary intake of magnesium is associated with an increased incidence of asthmatic symptoms, wheezing and reduced lung function.

Magnesium sulfate has been shown to inhibit smooth muscle contraction, decrease histamine release from mast cells, and inhibit acetylcholine release.

Sodium[17]:

Important for acid-base balance.

Required for normal cell permeability.

Sodium deficiency causes muscle cramps.

Iron[48]:

It is a component of haem which is required for the formation of haemoglobin. The function of hemoglobin is to carry the respiratory gases, oxygen and carbondioxide.

Alkaloids[49]:

Alkaloids have the property of dilating the bronchial tubes allowing an increase in breathing. So, the alkaloids containing plants in Singathi chooranam may help to treat the Asthma.

Tannic acid[50]:

It decreases the bronchial hyperreactivity, hence it helps to treat the Asthma.

Amino acid[51]:

It is an important antioxidant that may influence susceptibility to Asthma.

Toxicological studies:

From the results obtained in this study, no toxic effect was observed upto 400mg/kg of Singathi Chooranam via oral route over a period of 28 days. So, it can be concluded that the Singathi Chooranam can be prescribed for therapeutic use in human with the dosage recommendations of upto 400mg/kg. body weight p.o.

Pharmacological studies:

Based on the results of the above study, it can be concluded that the Singathi chooranam possesses significant antihistaminic and bronchodilating activity.

Clinical observation:

From the clinical study, after the course of treatment, 85% of patients relieved from wheezing, 70% of patients relieved from dyspnoea, 60% of patients relieved from cough, 66.67% of patients relieved from expectoration, 88.23% of patients relieved from chest tightness and no adverse effects were observed during trial period.

16(80%) of the patients had significant improvement in the PEFR after treatment.

Bio-statistics:

Statistically, the paired 't' test shows statistical significance for the PEFR and symptoms before and after the treatment.($p<0.0001$)

Siddha aspect[19]:

கார்ப்பு சுவையானது கபத்தை சமப்படுத்தும் தன்மையுடையது.

கைப்பு சுவையானது பித்தம், கபம் ஆகியவற்றின் விகற்பத்தை சாந்தி செய்யும்.

துவர்ப்பு சுவையானது கப பித்தத்தை போக்கும். வெப்பம் வீரியம் கபத்தை நீக்கும்.

கப குற்றத்தினால் ஏற்படும் இரைப்பு நோயினை கார்ப்பு சுவையுடைய மூலிகைகளான கண்டங்கத்திரி, சடாமாஞ்சில், சுக்கு, கடுக்காய், மிளகு, கைப்பு சுவையுடைய மூலிகைகளான கண்டுபாரங்கி, மிளகு, துவர்ப்பு சுவையுடைய மூலிகைகளான கர்க்கடகசிங்கி, கண்டுபாரங்கி, தான்றிக்காய் முதலிய சுவையுடைய மூலிகைகள் சிங்காதி சூரணத்தில் சேர்ந்துள்ளதால், இதனால் மருத்துவம் செய்ய நல்ல பலனை அளிக்கின்றது.

SUMMARY

The drug Singathi Chooranam was selected to evaluate the anti-histaminic and bronchodilator activity in the management of Eraippu (Bronchial asthma).

The literary evidence from Agathiyar mani ennum Vaidhiya Chinthamani Venba 4000–part 1 strongly support the antihistaminic, bronchodilator activity of the drug.

The qualitative and quantitative analysis were done at Biochemistry lab, National Institute of Siddha and Sri Ramachandra University, Chennai respectively. The biochemical analysis of the drug reveals the presence of sodium, magnesium, iron, chloride, alkaloids, tannic acid and amino acid. The results ensure the anti-histaminic and bronchodilator activity of the Singathi Chooranam was due to the presence of active phytoconstituents of the drug.

High Performance Thin Layer Chromatography and Atomic Absorption Spectrophotometer was done at Sri Ramachandra University, Chennai.

The pre-clinical evaluation (acute & repeated oral toxicity study) of the drug was carried out as per OECD guideline in Vels college of pharmacy, Chennai. The result shows safety of the drug for human administration.

The Preclinical Pharmacological study was carried out in animal model in Vels college of pharmacy, Chennai. The result shows that the drug has significant anti-histaminic and bronchodilator effect.

As per the Siddha literature and modern science reviews and research articles, the trial drug has potent anti-histaminic and bronchodilator effect.

20 Patients were recruited for clinical trial. The trial drug Singathi chooranam at the dose of 1 gm, b.i.d was given to the patient for 7 days and patients were asked to visit op once in 7 days for 30 days. Clinical assessment and prognosis was noted at each visit.

From the clinical study, after the course of treatment, 16(80%) of the patients had significant improvement in the PEFR after treatment.

85% of patients relieved from wheezing, 70% of patients relieved from dyspnoea, 60% of patients relieved from cough, 66.67% of patients relieved from expectoration,

88.23% of patients relieved from chest tightness and no adverse effects were observed during trial period.

Statistically, the paired 't' test shows statistical significance for the PEFR and associated symptoms before and after the treatment.($p<0.0001$)

The drug Singathi chooranam has

- Anti-histaminic Activity
- Bronchodilator Activity
- No side effects
- No undoing effects
- Encouraging clinical results.

From the clinical and the statistical analysis, it is proved that the drug Singathi Chooranam is statistically significant on antihistaminic and bronchodilator activity in the management of Eraippu (Bronchial asthma).

CONCLUSION

The literature and research journal review of the plant shows that it has anti-histaminic and bronchodilator activity.

The safety studies (acute toxicity and repeated oral toxicity studies) conducted revealed that the trial drug Singathi Chooranam is safe. There were no abnormalities found in blood investigation and histo-pathological examination. Hence it can be reasonably assumed that the drug is safe for human use.

The pharmacological study conducted in animal model shows significant Anti histaminic and Bronchodilator activity.

Clinical study reveals the therapeutic efficacy of the trial drug by showing, reduction in AEC levels and improvement in PEFR level significantly. There was improvement in other clinical symptoms after treatment.

There were no adverse reactions complained during the clinical trial.

Hence, the drug SINGATHI CHOORANAM can be used in the management of Eraippu (Bronchial Asthma)

TABLES FOR TRAIL DRUG-1 VALENDRAPHOLA CHOORANAM

TRIAL DRUG I, TABLE 1: QUALITATIVE ANALYSIS

S.NO	PARAMETERS	RESULTS
1.	Phosphate	Absent
2.	Sulphate	Absent
3.	Magnesium	Absent
4.	Iron	Present
5	Aminoacids	Present
6.	Starch	Absent
7.	Flavonoids	Absent
8.	Proteins	Present
9.	Tannic acid	Absent
10.	Glycosides	Absent

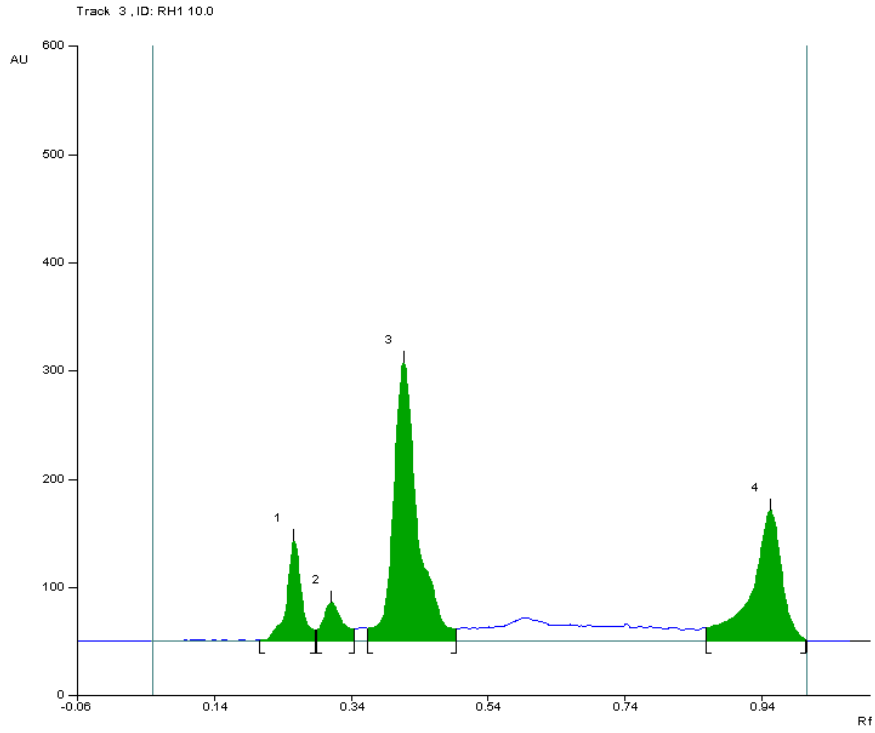
TRIAL DRUG I, TABLE 2: PHYSICAL PROPERTIES

S.NO	Characteristic test	Results
1.	Ph	4.6
2.	Ash Value	0.94
3.	Water soluble ash	0.03
4.	Acid insoluble ash	0.05

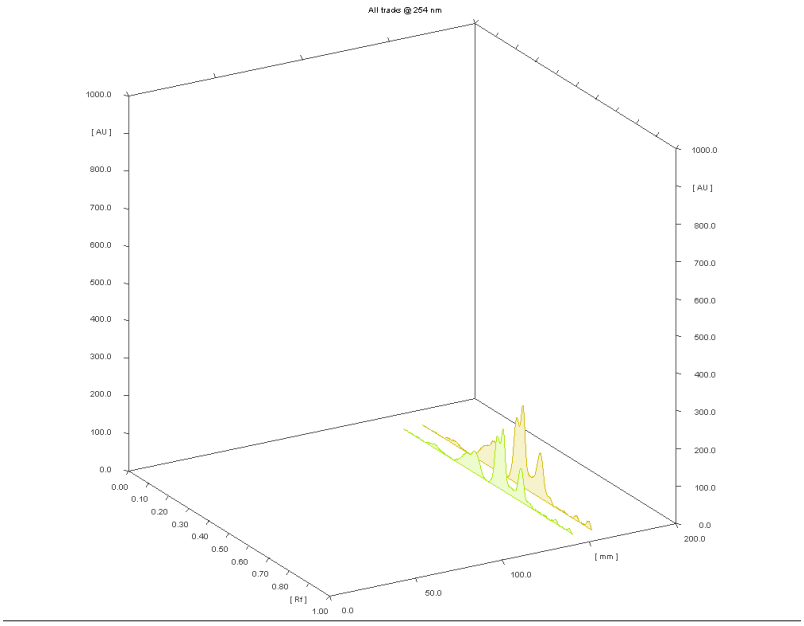
TRIAL DRUG I, TABLE 3: Preliminary acid, basic radicals screening of Valendraphola chooranam.

S.No.	Constituents	VC
1.	Calcium	+
2.	Iron (Ferric)	+
3.	Iron (Ferrous)	+
4.	Chloride	−
5.	Phosphate	−
6.	Potassium	−
7.	Sodium	−
8.	Sulphate	−

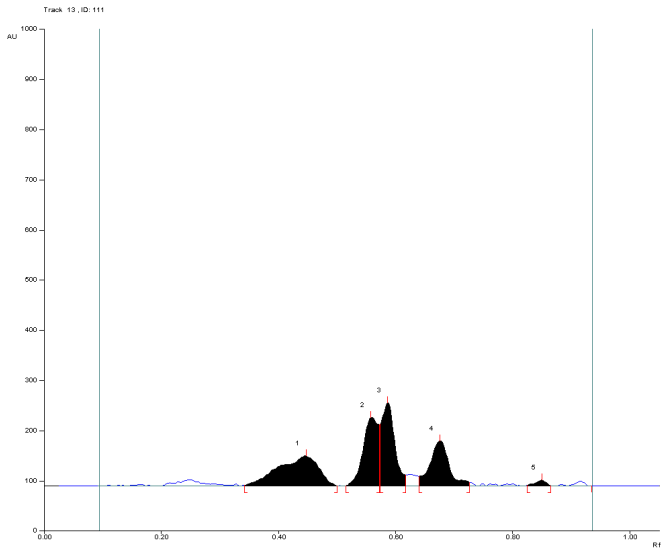
GRAPH 1: HPTLC Profile



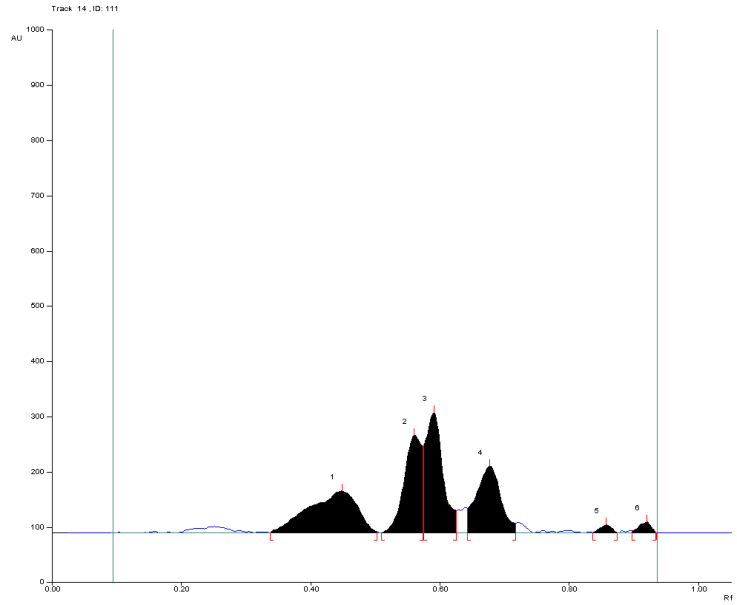
254nm



254nm 3D display

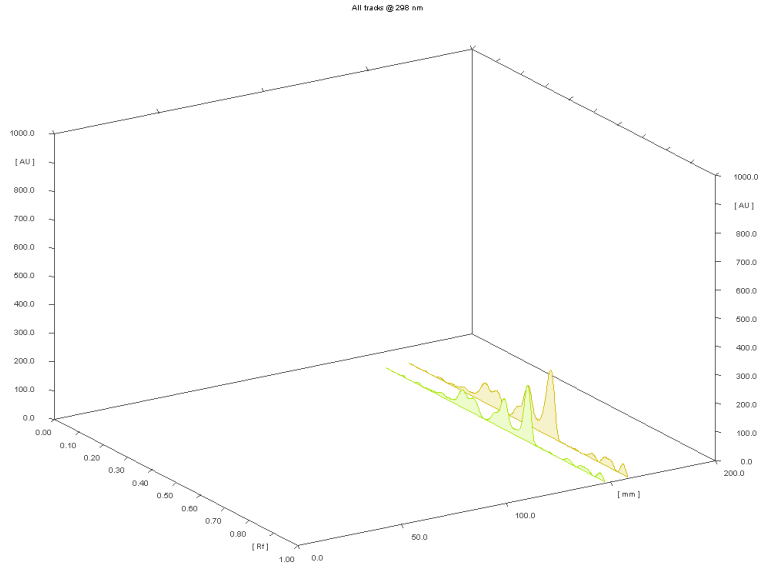


5µl (254nm)

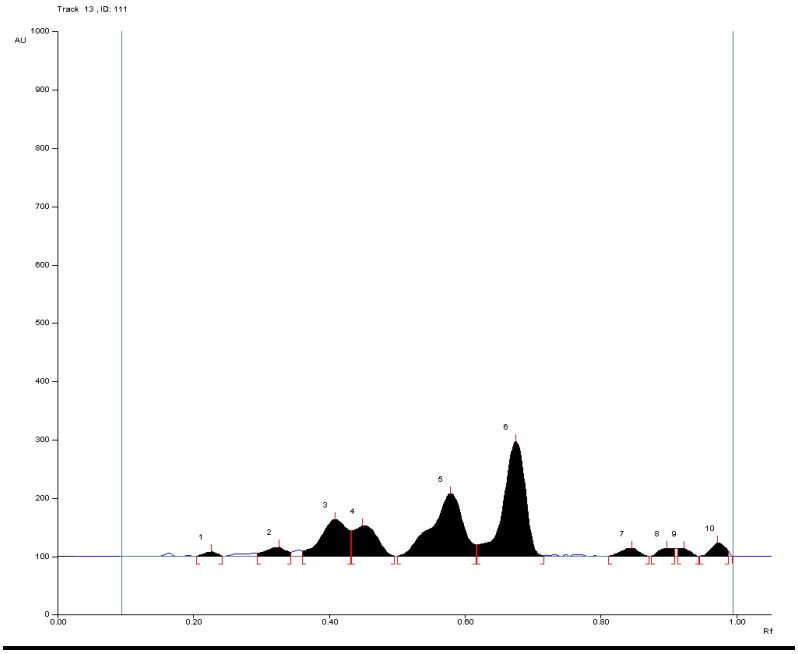


10 µl (254nm)

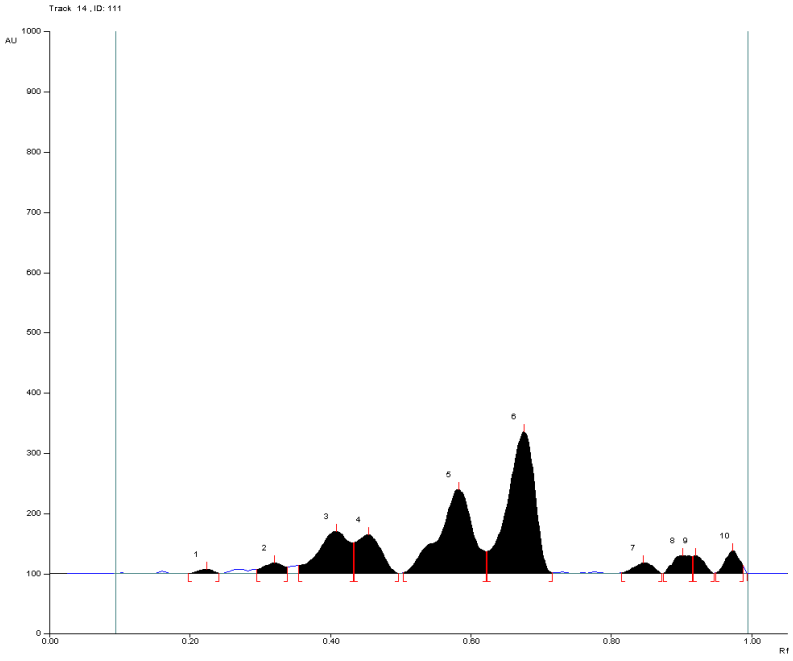
298nm



298 nm 3D display

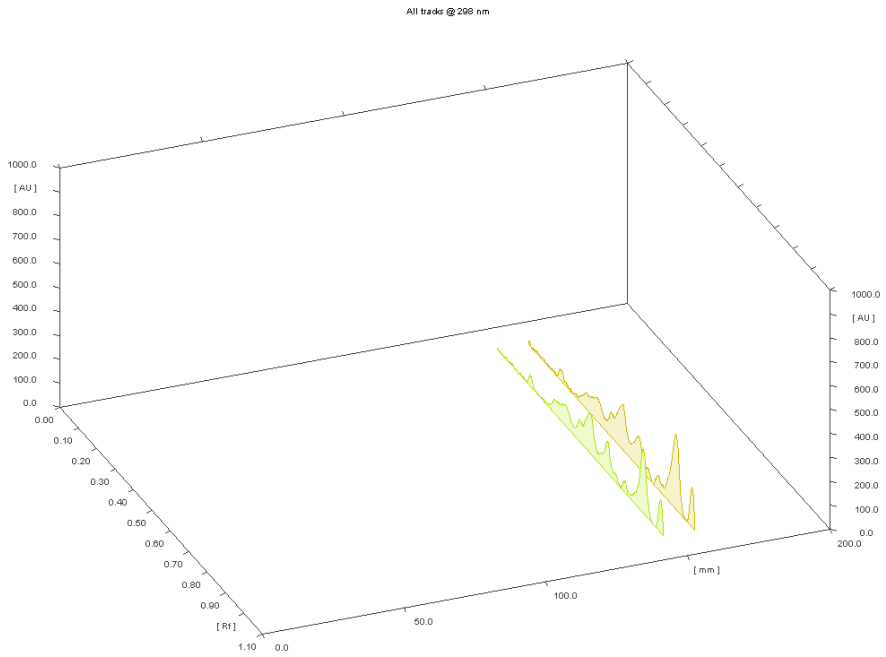


5µl (298nm)

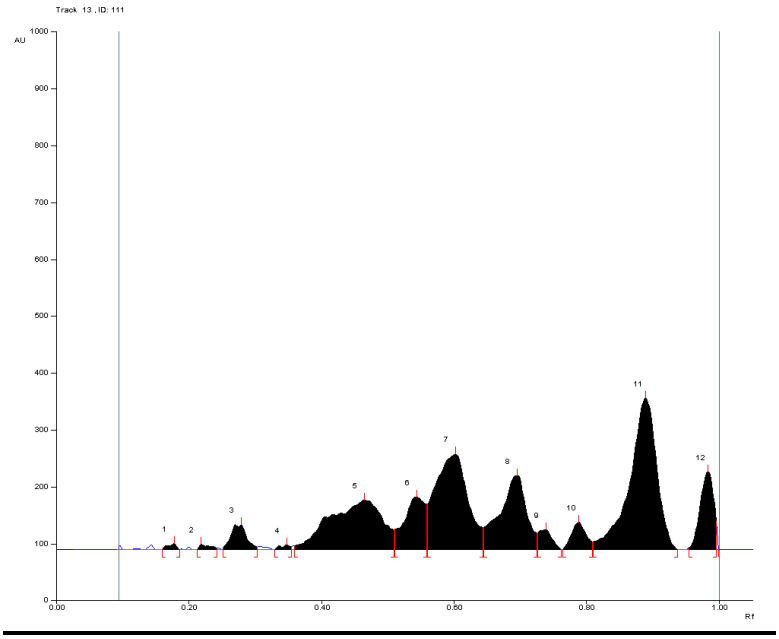


10µl (298nm)

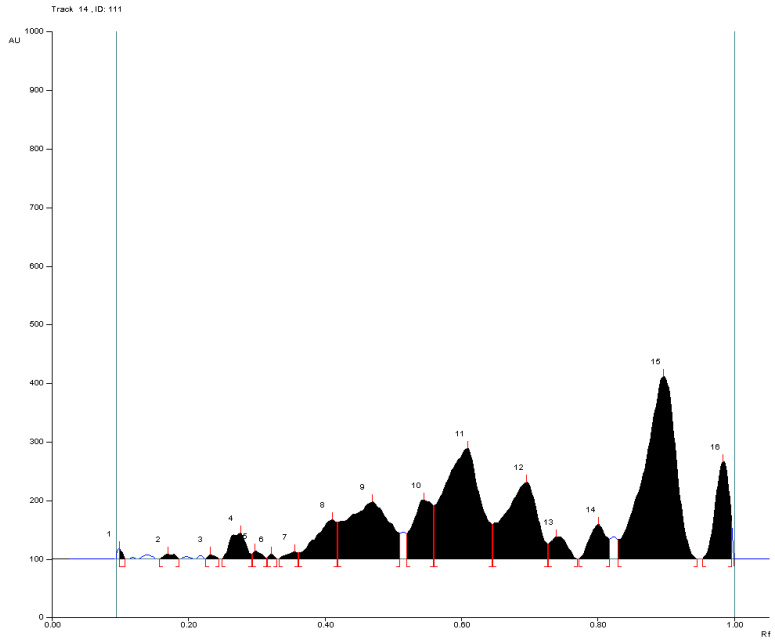
Derivatisation (298nm)



298 nm 3D display

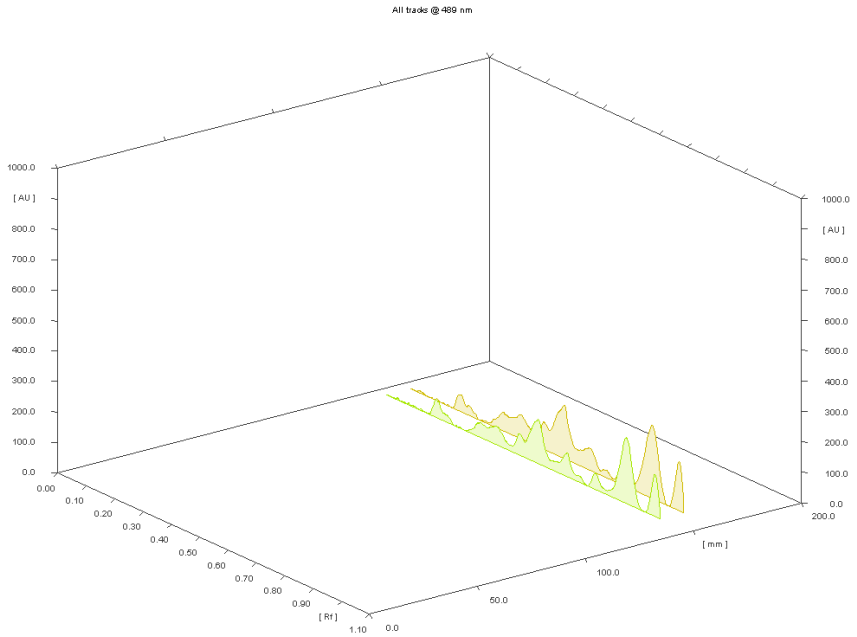


Derivatisation 5µl (298nm)

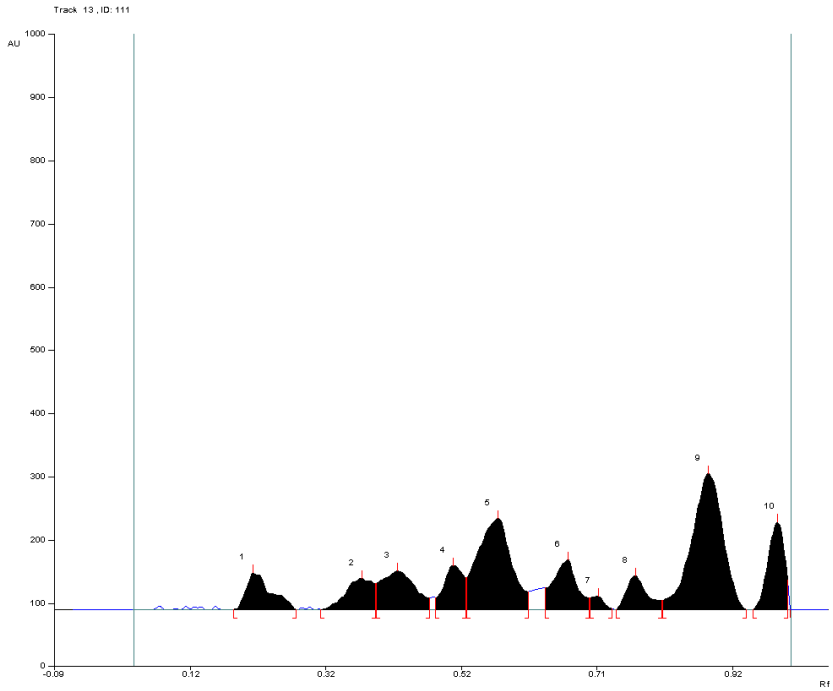


Derivatisation 10µl (298nm)

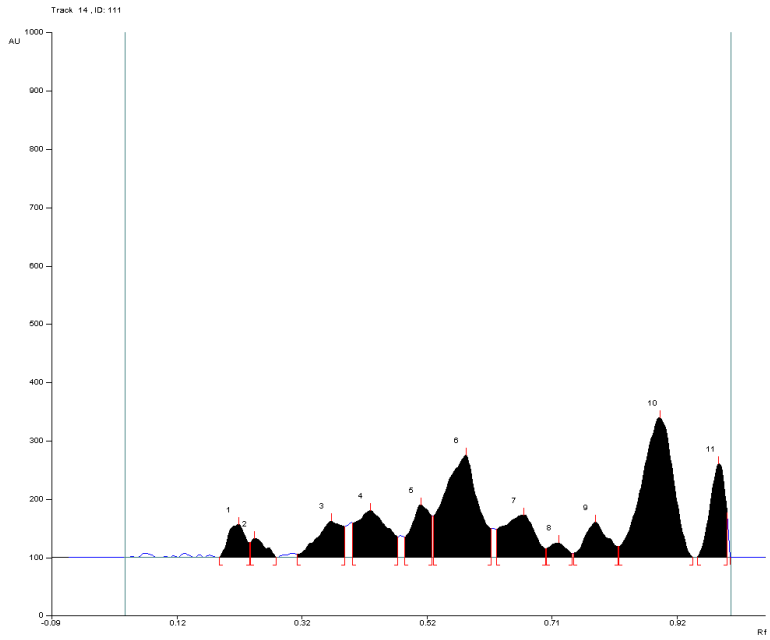
Derivatisation (498nm)



489 nm 3D display

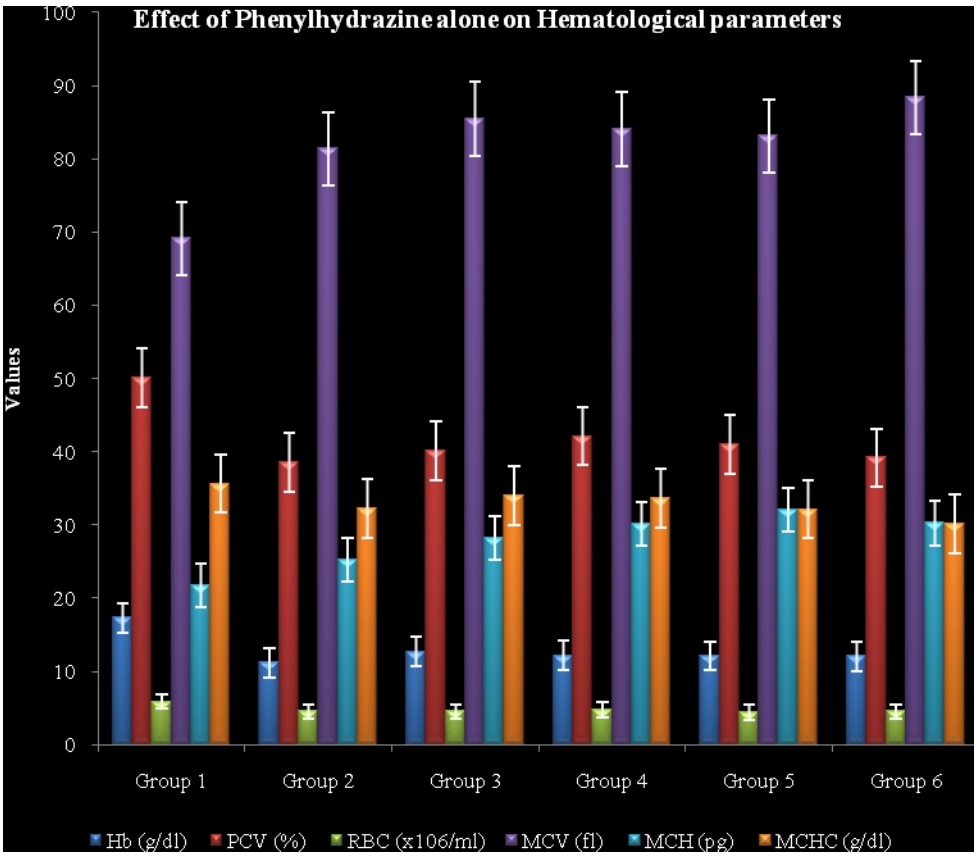


Derivatisation 5µl (498nm)

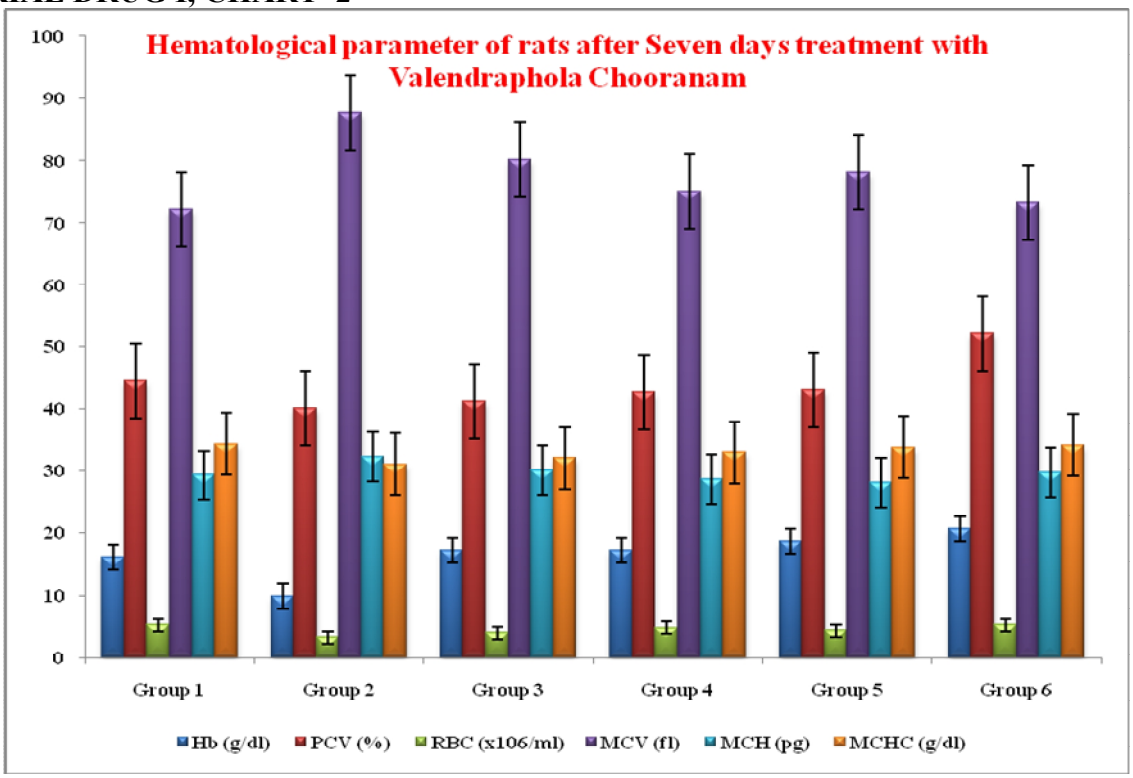


Derivatisation 10µl (498nm)

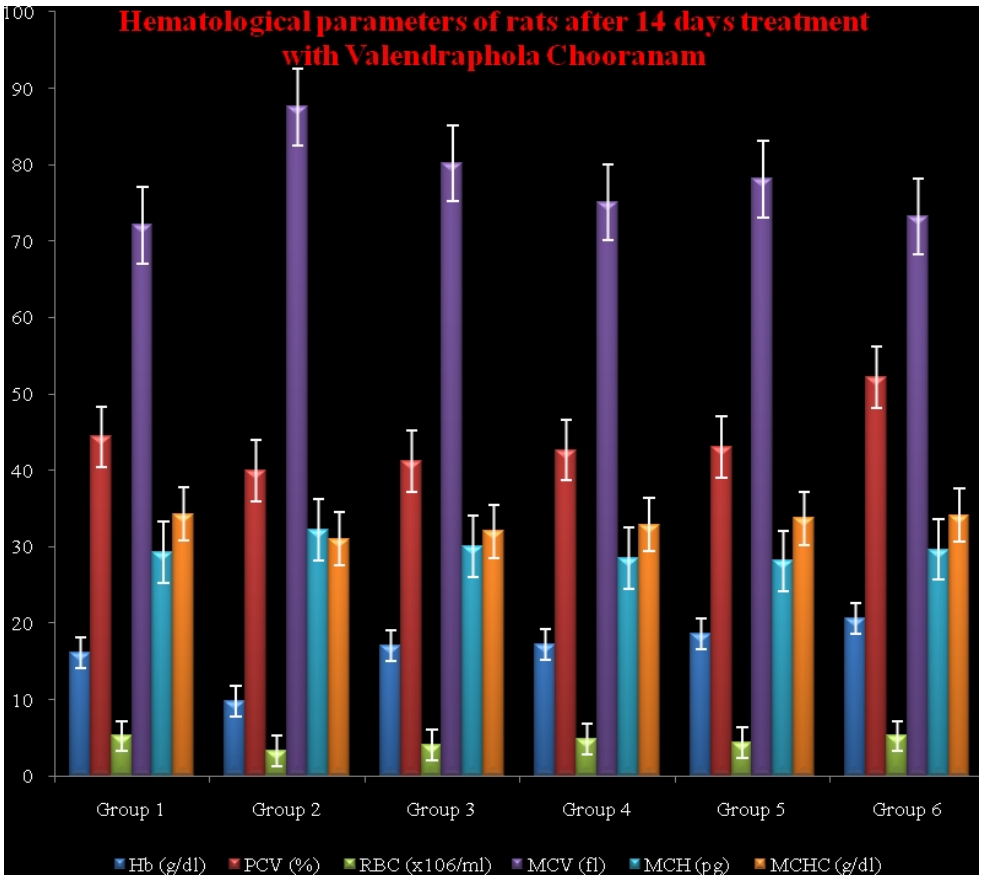
TRIAL DRUG I, CHART 1



TRIAL DRUG I, CHART 2



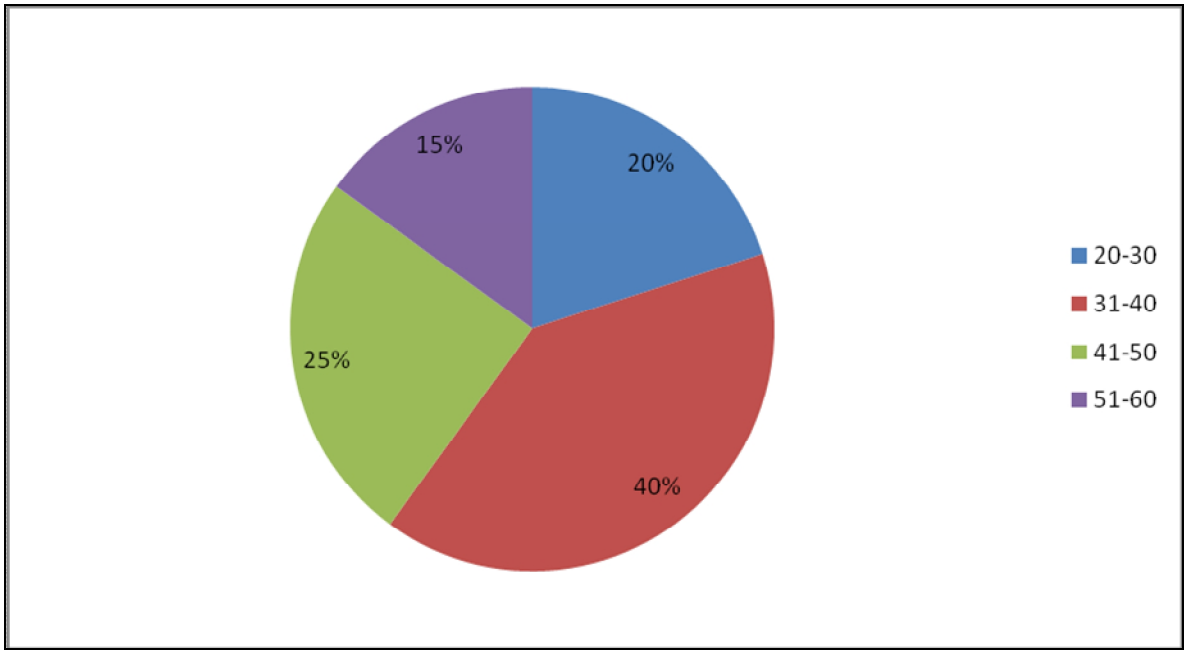
TRIAL DRUG I, CHART 3



CLINICAL OBSERVATION

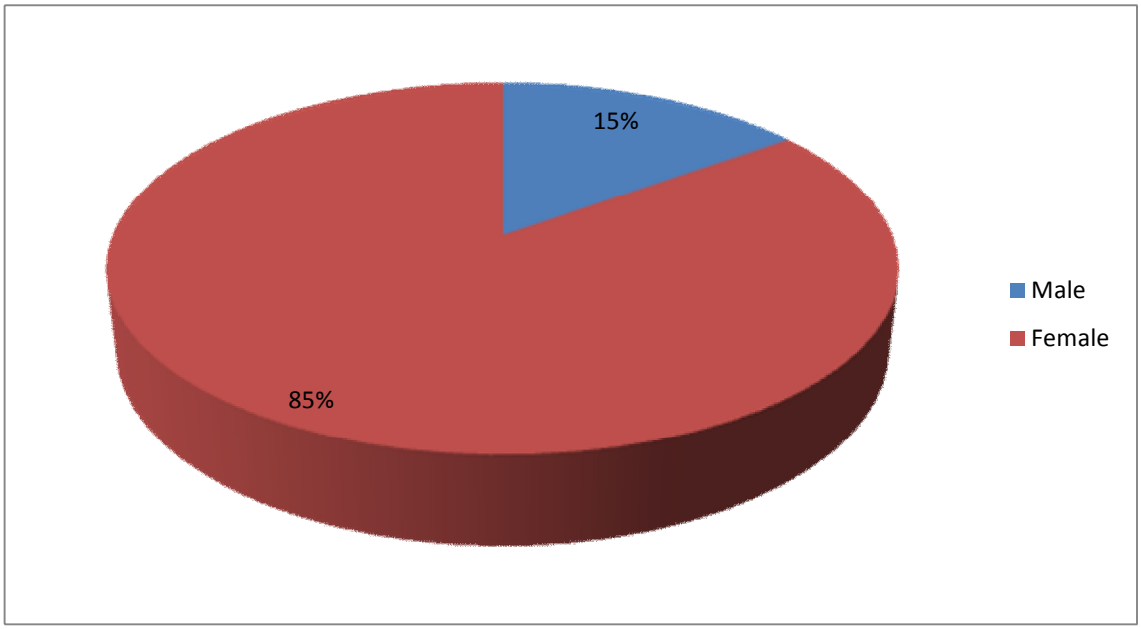
TRIAL DRUG I, TABLE 15: AGE DISTRIBUTION

S.NO	AGE(YEARS)	PERCENTAGE
1.	20-30	4(20%)
2.	31-40	8(40%)
3.	41-50	5(25%)
4.	51-60	3(15%)



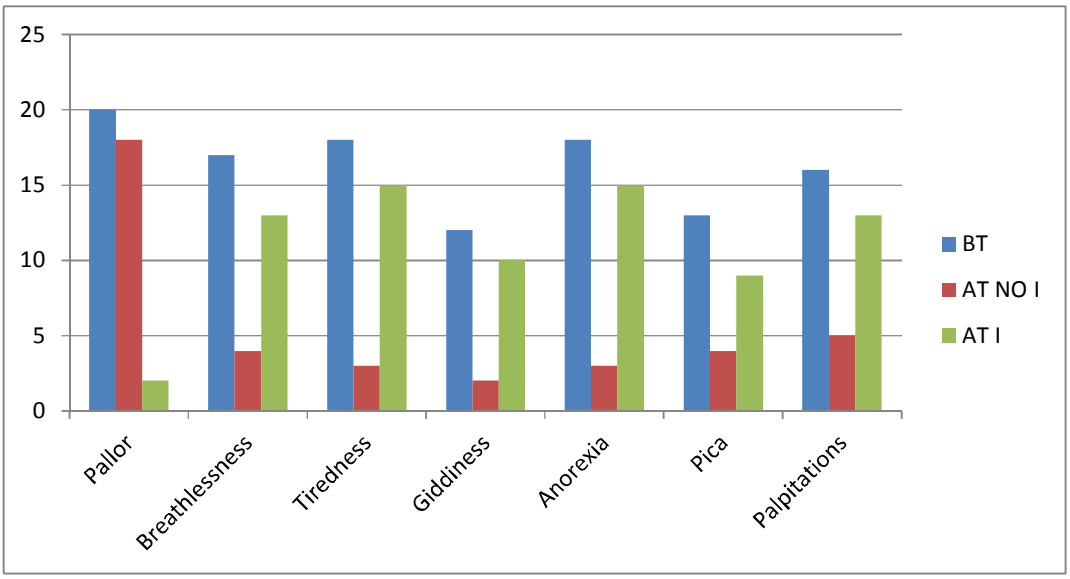
TRIAL DRUG I, TABLE 16: GENDER DISTRIBUTION

S.NO	GENDER	NO OF PATIENTS	PERCENTAGE
1.	MALE	3	15%
2.	FEMALE	17	85%



TRIAL DRUG I, TABLE 17: IMPROVEMENT SHOWING SIGNS AND SYMPTOMS BEFORE AND AFTER TREATMENT OF PANDU PATIENTS.

S.NO	SYMPTOMS	NO. OF PATIENTS WITH SYMPTOMS			
		BT	AT		Improvement Percentage
			No improvement	Improvement	
1.	Pallor	20(100%)	18(90%)	2(10%)	(10%)
2.	Breathlessness	17(85%)	4(23.53%)	13(76.47%)	(76.47%)
3.	Tiredness	18(90%)	3(16.67%)	15(83.33%)	(83.33%)
4.	Giddiness	12(60%)	2(16.67%)	10(83.33%)	(83.33%)
5.	Anorexia	18(90%)	3(16.67%)	15(83.33%)	(83.33%)
6.	Pica	13(65%)	4(30.77%)	9(69.23%)	(69.23%)
7.	Palpitations	16(80%)	3(18.75%)	13(81.25%)	(81.25%)



BT – Before Treatment
AT NOI – After Treatment No Improvement
AT I – After Treatment Improvement

IMPROVEMENT IN HB LEVELS

S.NO	OP/IP NO	AGE	SEX	BT HB	AT HB
1	C81998	52	F	9	9.7
2	C81264	39	F	6.4	7.8
3	C75674	55	F	9.3	10.6
4	C83901	25	F	8.4	9.1
5	C82169	45	M	10	10
6	C69781	36	F	7.4	8.9
7	C91254	39	F	9.2	11.1
8	C92794	33	F	8.6	11
9	C91728	27	F	7.2	8.2
10	C92876	40	F	8.2	8.9
11	C92882	51	F	7.1	7.1
12	C82212	28	M	8.6	9.8
13	C93377	37	F	7.6	7.6
14	C91376	37	F	8.3	9.3
15	C94453	49	F	9.2	9.2
16	C89619	38	F	9.6	10.8
17	C94419	46	F	9	10.4
S18	4248	48	F	8.5	10
19	D000707	47	M	8.9	10
20	C90994	28	F	6.1	7.6

STATISTICAL ANALYSIS

All collected data were entered into MS Excel software using different columns as variables and rows as patients. SPSS software was used to perform statistical analysis. Basic descriptive statistics include frequency distributions and cross-tabulations were performed. The quantity variables were expressed as Mean ± Standard Deviation and qualitative data as percentage. A probability value of <0.05 was considered to indicate as statistical significance. Paired 't' test was performed for determining the significance between before and after treatment.

Paired t test for Symptoms before and after treatment:

Variable	Obs	Mean	Std.dev	t.value	P value
Before symptoms	20	5.10	1.689	10.353	P<0.0001
After symptoms	20	1.85	1.631		

For Symptoms, the mean±standard deviation before treatment is 5.10±1.689 and after treatment is 1.85±1.631, which is statistically significant($p<0.0001$).

Paired t test for Hb before and after treatment:

Variable	Obs	Mean	Std.dev	t.value	P value
Before treatment	20	8.33	1.0553	-6.945	P<0.0001
After treatment	20	9.355	1.2041		

For Hb, the mean±standard deviation before treatment is 8.33±1.0553 and after treatment is 9.355±1.2041, which is statistically significant($p<0.0001$).

TOXICOLOGY TABLES

TRIAL DRUG I, TABLE 4: Dose finding experiment and its behavioural Signs of Toxicity

No	Dose mg/kg	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1	2000	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2	5000	+	-	-	+	-	+	+	-	-	-	-	-	+	-	-	-	-	-	-	-

1. Alertness 2. Aggressiveness 3. Pile erection 4. Grooming 5. Gripping 6. Touch Response 7. Decreased Motor Activity 8. Tremors 9. Convulsions 10. Muscle Spasm 11. Catatonia 12. Muscle relaxant 13. Hypnosis 14. Analgesia 15.Lacrimation 16. Exophthalmos 17. Diarrhoea 18. Writhing 19. Respiration 20. Mortality

TRIAL DRUG I, TABLE 5. Body wt (g) of rats exposed to *Valendraphola Chooranam* for 28days.

Dose (mg/kg/day)	*Days*				
	1	7	14	21	28
Control	122.65±4.22	124.13±5.10	126.10±4.12	128.52±7.02	128.62±5.21
100	124.47±5.10	125.22±5.12	126.12±4.70	125.22±5.88	126.10±5.52
250	124.18±4.42	128.17±5.75	124.18±5.12	125.18±6.00	126.17±6.10
500	121.15±4.21	125.23±5.38	128.16±4.71	128.02±6.20	128.10±6.22

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$; Vs Control N=6.

TRIAL DRUG I, TABLE 6. Food intake of rats exposed to *Valendraphola Chooranam* for 28days.

Dose (mg/kg/day)	Days (gms/rats)				
	1	**7**	**14**	**21**	**28**
Control	47.05±2.32	45.12±2.52	54.41±2.31	55.11±2.66	57.52±3.16
100	44.27±2.28	45.34±2.40	52.40±2.88	50.11±2.26	48.10±3.22
250	45.13±2.14	42.82±2.54	44.42±2.64*	45.32±3.46	46.48±3.00
500	44.41±2.45	45.48±2.55	42.32±2.57*	45.10±2.60*	45.44±3.18*

Values are mean ± S.E.M. (Dunnet 't' test). ^{ns}P>0.05; Vs Control N=6.

TRIAL DRUG I, TABLE 7. Water (ml/day) intake of rats exposed to *Valendraphola Chooranam* for 28days.

Dose (mg/kg/day)	Days(ml/rat)				
	1	**7**	**14**	**21**	**28**
Control	50.22±2.45	52.44±3.12	55.28±3.10	52.14±3.18	51.12±3.45
100	50.10±2.40	50.96±3.04	45.23±3.46	46.00±3.00	45.55±2.51
250	47.27±2.84	40.18±3.34*	40.87±3.28*	42.32±2.48	42.10±3.20
500	51.14±3.45	52.44±3.15	50.20±3.41	50.13±3.12	53.22±3.44

Values are mean ± S.E.M. (Dunnet't' test). *P<0.05; N=6.

TRIAL DRUG I, TABLE 8. Hematological parameters after 28days treatment with *Valendraphola Chooranam*.

Parameter	Control	100 mg/kg	250 mg/kg	500 mg/kg
RBC (millions/cu.mm)	5.13±0.41	5.15±0.41	5.26±0.32	5.32±0.41
Hb (g/dl)	13.01±0.28	13.83±0.98	14.0±1.1	14.12±1.0
PCV (%)	42.11±1.05	44.5±4.1	44.0±2.5	45±1.1
WBC(cells/cu.mm)	7375±440.17	8050±330.9	8325±417.3	8633±513.3
Neutrophil (%)	45.22±4.10	42.42 ±2.5	45.24±3.0	45.27±3.2
Lymphocytes (%)	40.22±2.26	42.10±2.13	42.15±2.40	44.10±4.1
Eosinophil's (%)	4.0±0.40	3.0±0.56	4.1±0.44	5.10±0.43
Monocytes (%)	3.0±0.22	4.0±0.24*	4.0±0.3*	3.2±0.2
Basophils (%)	0±0	0±0	0±0	0±0
Platelets (10^5 cells/cu.mm)	1.52±0.05	1.86±0.09	1.89±0.12	2.1±0.14**
MCV(Fl)	75.5±2.2	67.2±1.4	73.1±2.3	74.4±3.5
MCHC (pg)	29.5±2.5	28.41±2.4	29.2±2.1	30.00±2.5

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; **P<0.01. N=6.

TRIAL DRUG I, TABLE 9. Effect of *Valendraphola Chooranam* biochemical (LFT, RFT, Lipid Profile) Parameters

Parameter	Control	100 mg/kg	250 mg/kg	500 mg/kg
Glucose (mg/dL)	85.04±5.17	78.66±5.50	76.20±7.25	74.16±6.88
Total Bilirubin (mg/dL)	0.213±0.05	0.214±0.05	0.212±0.05	0.210±0.06
Bilirubin direct (mg/dL)	0.1±0.04	0.1±0.05	0.1±0.04	0.1±0.05
Creatinine (mg/dL)	0.90±0.05	0.88±0.05	0.91±0.03	0.93±0.03
BUN (mg/dL)	18.05±1.56	18.00±1.29	17.26±1.73	17.14±1.64
AST (IU/L)	132.4±7.32	127.2±6.12	126.1±6.10	124.2±5.18
ALT (IU/L)	36.10±3.11	34.10±2.46	35.52±2.55	34.42±2.40
ALP (IU/L)	75.32±4.43	70.16±4.12	66.30±4.36	67.44±4.22
Total cholestrol (mg/dL)	56.86±5.77	57.61±5.68	55.84±5.21	59.61±4.88
Total protein (g/dL)	8.30±0.28	7.44±0.24	7.58±0.75	7.56±0.76
Albumin (g/dL)	2.62±0.05	2.64±0.06	2.72±0.06	2.80±0.05
Urea(mg/dL)	55.32±1.60	54.41±3.50	55.12±2.38	54.75±2.16
Uric acid (mg/dL)	1.6±0.12	1.6±0.18	1.6±0.16	1.5±0.14
Na m.mol	142.70±5.15	141.2±5.00	141.45±5.20	141.22±5.12
K m.mol	20.11±2.47	19.88±1.72	20.10±1.62	20.16±1.72
Cl m.mol	102.16±4.11	102.33±5.14	104.44±5.82	100.04±5.10
HDL(mg/dL)	13.00±1.14	13.31±1.45	13.10±1.42	13.46±2.00
LDL(mg/dL)	43.12±2.23	44.55±3.54	42.21±3.23	43.10±3.24
VLDL(mg/dl)	15.42±2.55	15.98±2.65	16.02±2.34	15.98±2.20
Triglycerides (mg/dl)	85.24±3.00	85.26±2.22	86.23±3.24	88.36±2.72

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$; Vs Control N=6.

TRIAL DRUG I, TABLE-10 Urine Analysis

Parameters	Control	100 mg/kg	250 mg/kg	500 mg/kg
Colour	Yellow	Yellow	Yellow	Yellow
Transparency	Clear	Slightly turbid	Slightly cloudy	Slightly turbid
Specific gravity	1.010	1.010	1.010	1.010
PH	>7.2	>8.0	>8.0	>9.0
Protein	Nil	3+	3+	3+
Glucose	Nil	Nil	Nil	Nil
Bilirubin	-ve	-ve	-ve	-ve
Ketones	-ve	+ve	+ve	+ve
Blood	Absent	Absent	Absent	Absent
Urobilinogen	Normal	Abnormal	Abnormal	Abnormal
Pus cells	0-cells/HPF	1-cell/HPF	2-cells/HPF	1-cell/HPF
RBCs	Nil	Nil	0-1cells/HPF	Nil
Epithelial cells	Nil	1-cell/HPF	Nil	1-cell/HPF
Crystals	Nil	Nil	Nil	Nil
Casts	Nil	Nil	Nil	Nil
Others	Bacteria seen	Bacteria seen	Bacteria seen	Bacteria seen

TRIAL DRUG I, TABLE 11. Effect of *Valendraphola Chooranam* on organ weight

Dose (mg/kg)	Control	100 mg/kg	250 mg/kg	500 mg/kg
Heart(g)	0.70±0.05	0.67±0.11	0.66±0.09	0.69±0.04
Liver(g)	4.42±0.42	4.29±0.39	4.82±0.62	4.77±0.55
Lung(g)	0.71±0.04	0.72±0.05	0.72±0.05	0.69±0.4
Spleen(g)	0.68±0.04	0.69±0.04	0.71±0.05	0.68±0.05
Kidney(g)	1.09±0.3	1.01±0.04	1.96±0.2**	1.42±0.05
Testis(g)	0.92±0.03	0.91±0.05	0.90±0.04	0.91±0.03
Ovary(g)	0.04±0.01	0.04±0.02	0.04±0.01	0.04±0.02
Brain	0.69±0.06	0.67±0.04	0.68±0.03	0.67±0.05
Pancreas	1.44±0.08	1.48±0.09	1.50±0.10	1.51±0.12
Uterus	0.78±0.08	0.82±0.09	0.81±0.06	0.82±0.05

Values are mean ± S.E.M. (Dunnet’t' test). *P<0.05; **P<0.01 Vs Control N=6.

PHARMACOLOGICAL TABLES

TRIAL DRUG I, TABLE 12: Effect of Phenylhydrazine (10mg/kg, p.o. daily for 7 days) alone on Hematological parameters.

Parameters	Group 1 (Normal)	Group 2 (Anemic)	Group 3 (Anemic)	Group 4 (Anemic)	Group 5 (Anemic)	Group 6 (Anemic)
Hb (g/dl)	17.31 ± 0.46	11.2±0.27**	12.65±0.25**	12.14±0.38**	12.11±0.28**	12.05 ± 0.30**
PCV (%)	50.12 ±1.51	38.55 ± 2.39**	40.12 ± 1.53*	42.12 ± 2.34	41.05 ± 2.04*	39.16 ± 2.72**
RBC (x10^6/ml)	5.88 ± 0.17	4.50 ± 0.28**	4.53 ± 0.24**	4.75 ± 0.20*	4.44 ± 0.30**	4.52 ± 0.34**
MCV (fl)	69.15 ± 2.67	81.35±3.4	85.46±6.11*	84.05±3.12	83.16±5.44	88.41± 3.61*
MCH (pg)	21.74 ± 1.44	25.28±1.31	28.19±1.65*	30.12±1.10**	32.10±0.84**	30.24 ± 2.40**
MCHC (g/dl)	35.64 ± 0.38	32.20±0.5	34.02±0.74	33.62±1.46	32.14±2.86	30.09 ± 2.22

Values are ± S.E.M. (Dunnet't' test). *P<0.05; **P<0.01 Vs Control N=6.

TRIAL DRUG I, TABLE 13:Hematological parameter of rats after Seven days treatment with Valendraphola Chooranam.

Parameters	Group 1 (Normal)	Group 2 (Anemic control)	Group 3 (100 mg/kg)	Group 4 (200 mg/kg)	Group 5 (400 mg/kg)	Group 6 (Heamatinic syrup)
Hb (g/dl)	17.11 ±1.26**	11.31±1.1	13.64±0.48	14.55±1.22	15.64 ± 0.59*	19.54 ±1.41**
PCV (%)	51.04 ±1.34**	40.10±2.6	43.21±2.29	45.25±2.37	49.46±2.48*	54.18 ±2.48**
RBC (x10^6/ml)	5.16±0.21	4.12±0.28	5.16±0.24	5.28±0.18	5.11±0.21	6.55±1.00**
MCV (fl)	75.17±2.36	74.2±2.5	78.20±1.8	75.4±2.6	76.30±4.1	75.33±2.52
MCH (pg)	22.10±1.48*	28.1±1.42	22.24±1.5*	23.0±1.4	23.05±2.0	22.26±1.34*
MCHC (g/dl)	34.24±1.28	31.15±1.18	28.64±1.8	29.79±1.5	31.2±1.7	33.17±1.4

Values are mean ± S.E.M. (Dunnet't' test). *P<0.05; **P<0.01 Vs Control N=6.

TRIAL DRUG I, TABLE 14:Hematological parameters of rats after 14 days treatment with Valendraphola Chooranam.

Parameters	Group 1 (Normal)	Group 2 (Anemic control)	Group 3 (100 mg/kg)	Group 4 (200 mg/kg)	Group 5 (400 mg/kg)	Group 6 (Heamatinic syrup)
Hb (g/dl)	16.12 ±1.47**	9.78 ± 1.17	17.10 ± 0.68*	17.15 ± 0.56**	18.56 ± 0.54**	20.64 ±1.38**
PCV (%)	44.38 ± 1.31	40.00±1.87	41.14±3.02	42.64±2.2	43.02±1.9	52.17 ± 1.82**
RBC ($x10^6$/ml)	5.22±0.26**	3.28±0.50	4.00±0.37	4.81±0.34*	4.38±0.28	5.22±0.52**
MCV (fl)	72.10±2.08**	87.56±2.85	80.16±2.40	75.10±1.61**	78.11±2.36*	73.21±2.46**
MCH (pg)	29.24±2.85	32.19±2.24	30.00±1.48	28.54±1.74	28.13±1.27	29.65±2.17
MCHC (g/dl)	34.26±2.88	31.05±1.34	32.02±1.33	32.89±0.68	33.74±2.58	34.10±3.02

Values are mean ± S.E.M. (Dunnet't' test). *P<0.05; **P<0.01 Vs Control N=6.

SPECIFIC INVESTIGATIONS FOR PANDU

S.NO	OP/IP NO	AGE	SEX	HB [gm/dl]		RBC [million/cu.mm]		PCV [%]		MCV [fL]		MCH [pg]		MCHC [gm/dl]		HM		ESR[mm]			
																		½ Hr		1 Hr	
				BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT
1	C81998	52	F	9	9.7	5	5	31	31	61.8	61.3	18.5	19.2	30	30.1	HM	HM	10	4	16	8
2	C81264	39	F	6.4	7.8	3.9	4	23.6	25	59.6	58.2	16.2	16.2	27.1	27.8	HM	HM	14	2	20	6
3	C75674	55	F	9.3	11	4.7	5	31	33.4	65	65.9	19.5	20.9	30	31.7	HM	N	12	6	32	12
4	C83901	25	F	8.4	9.1	4.6	4.9	28.8	30.7	61.4	61.6	17.9	18.3	29.2	29.6	HM	HM	10	4	14	8
5	C82169	45	M	10	10	5.2	5.3	38.4	38.5	80.2	80.1	30.1	30.1	34.3	34.3	HM	HM	8	6	20	18
6	C69781	36	F	7.4	8.9	4.3	4.8	26.1	28.6	60.1	60.3	17.1	17.1	28.4	28.6	HM	HM	6	2	14	10
7	C91254	39	F	9.2	11	3.3	3.9	28.2	30.9	98	98.1	30	30.6	34.7	35.1	HM	N	4	4	8	6
8	C92794	33	F	8.6	11	3.8	4.7	25.8	30.3	88.3	88.2	29.7	29.7	31	31.2	HM	N	12	10	24	20
9	C91728	27	F	7.2	8.2	4.1	4.4	25.1	25.9	60.6	60.1	17.4	16.7	28.7	27.8	HM	HM	4	6	8	12
10	C92876	40	F	8.2	8.9	4.4	4.6	28.3	30	63.7	64.7	18.5	19.2	29	29.7	HM	HM	4	2	10	8
11	C92882	51	F	7.1	7.1	4.4	4.4	26.8	26.8	59.7	59.5	15.8	15.8	26.5	26.5	HM	HM	4	4	8	10
12	C82212	28	M	8.6	9.8	4.7	5	29.4	32.5	72.2	72.4	20.2	20.4	28.4	28.6	HM	HM	10	6	18	12
13	C93377	37	F	7.6	7.6	4.3	4.4	25.1	25.1	57.2	57.2	14.4	14.5	25.1	25.3	HM	HM	20	14	42	22
14	C91376	37	F	8.3	9.3	4.6	4.8	28.5	29.3	61	61.8	17.8	18	29.1	29.9	HM	HM	16	6	22	12
15	C94453	49	F	9.2	9.2	4.7	4.6	30.4	30.4	65.1	65.1	19.6	19.6	30.1	29.9	HM	HM	6	2	20	4
16	C89619	38	F	9.6	11	4	4.4	31.3	32.2	67.5	67.9	20.7	21	30.7	30.9	HM	HM	8	4	16	12
17	C94419	46	F	9	10	4.4	4.7	31.1	31.9	67.2	67.3	19.9	20	29.6	29.6	HM	HM	4	4	12	10
18	4248	48	F	8.5	10	3.7	3.9	32.2	30.4	79.5	77.6	25.7	25.5	32.3	32.9	HM	HM	22	12	62	26
19	D000707	47	M	8.9	10	5	5.4	28.3	32.5	67.6	67.9	20.3	20.4	29.4	29.6	HM	N	12	6	20	12
20	C90994	28	F	6.1	7.6	4.1	4.2	23.2	23.6	55.6	55.7	14.6	14.6	26.3	26.3	HM	HM	18	14	24	20

PANDU – BEFORE TREATMENT

S NO	OP/IP NO	AGE	SEX	TC [cu.mm]	DC				PLT [Lakhs/cu.mm]	BLOOD SUGAR [mg/dl]		URE [mg/dl]	CRE [mg/dl]	CHO [mg/dl]	LFT				CA[mg/dl]	PHO [mg/dl]	U.A [mg/dl]	URINE				
					p%	L%	E%	M %		F	PP				OT [Iu/L]	PT [Iu/L]	ALP [Iu/L]	TP [mg/dl]				ALB	SUGAR		DEP	
																							F	PP	PUS	EPI
1	C81998	52	F	7700	58	33	6	3	3.6	107	127	15	0.6	136	14	15	140	6.7	11.2	3.4	4.4	nil	nil	nil	1-2	1-2
2	C81264	39	F	6200	55	40	5	0	2.7	96	122	14	0.5	156	21	25	148	7.9	10.3	3.5	5	nil	nil	nil	2-3	2-3
3	C75674	55	F	7400	55	35	10	0	4.2	89	106	15	0.5	140	33	16	150	7.4	10.5	3.2	3.6	nil	nil	nil	2-3	2-3
4	C83901	25	F	6300	48	48	4	0	3.2	82	99	18	0.5	96	17	18	131	7	10.9	2.9	3.1	nil	nil	nil	6-8	3-4
5	C82169	45	M	9000	63	32	5	0	3.1	103	138	14	0.5	211	37	40	220	7.6	11.3	4	3.6	nil	nil	nil	2-4	1-2
6	C69781	36	F	4500	45	48	7	0	3.2	92	106	14	0.4	90	15	16	166	7.3	11.1	3.2	3.2	nil	nil	nil	2-3	2-3
7	C91254	39	F	4700	64	30	5	1	2	102	116	14	0.4	105	35	36	245	6.5	11	3.1	3	nil	nil	nil	1-2	1-2
8	C92794	33	F	8500	53	41	4	2	2.2	85	119	22	0.6	125	21	24	168	5.2	9.9	2.8	3.8	nil	nil	nil	1-3	2-4
9	C91728	27	F	8200	63	34	3	0	3.2	92	101	25	0.7	115	14	15	147	7.2	9.8	2.9	2.9	nil	nil	nil	3-4	4-5
10	C92876	40	F	6000	50	44	6	0	2.6	83	105	15	0.4	130	17	18	165	7.6	9.8	3	4	nil	nil	nil	2-3	1-2
11	C92882	51	F	8900	68	28	4	0	4.9	99	134	20	0.6	147	14	13	166	7.2	11.3	2.9	5.2	nil	nil	nil	1-2	1-2
12	C82212	28	M	9300	55	38	6	1	2.3	107	132	25	0.8	194	18	23	182	7	11.5	3	5.9	nil	nil	nil	2-4	1-2
13	C93377	37	F	5300	50	42	6	0	3.1	94	121	14	0.4	146	22	23	164	7.2	10.5	3	3.7	nil	nil	Nil	3-4	1-2
14	C91376	37	F	6500	53	42	4	1	4.3	94	104	17	0.5	128	10	11	130	7.6	11	3	4	nil	nil	Nil	1-2	2-3
15	C94453	49	F	4700	54	39	5	2	2.5	86	116	15	0.4	168	16	18	173	5,8	10.5	3.1	4.2	nil	nil	Nil	2-4	2-4
16	C89619	38	F	7700	58	40	2	0	2.9	103	127	14	0.4	153	16	20	186	5.9	10.8	3.2	3.4	nil	nil	Nil	1-2	3-5
17	C94419	46	F	7000	59	36	4	1	4	89	118	15	0.4	135	15	17	161	6.2	11.6	3.4	3.6	nil	nil	Nil	2-3	2-3
18	4248	48	F	10200	72	23	5	0	3.2	99	124	14	0.5	194	13	15	189	6.8	11.2	3.5	4.4	nil	nil	Nil	8-10	4-8
19	D000707	47	M	6400	70	27	3	0	3.1	98	120	20	0.6	145	15	17	163	5	10.5	3.2	3.4	nil	nil	Nil	4-8	3-6
20	C90994	28	F	7300	57	33	10	0	4	101	112	14	0.4	110	14	16	171	6.4	10.8	3.2	3.3	nil	nil	Nil	3-4	4-5

PANDU – AFTER TREATMENT

S.NO	OP/IP NO	AGE	SEX	TC [cu.mm]	DC				PLT [Lakhs/ cu.mm]	BLOOD SUGAR [mg/dl]		UREA [mg/dl]	CREAT [mg/dl]	CHOL [mg/dl]	LFT				CA [mg/dl]	PHOS [mg/dl]	U.A [mg/dl]	URINE				
					P%	L%	E%	M%		FBS	PPBS				SGOT [Iu/L]	SGPT [Iu/L]	ALP [Iu/L]	TP [mg/ dl]				ALB	SUGAR		DEP	
																							F	PP	PUS	EPI
1	C81998	52	F	7700	65	30	5	0	3.3	110	120	18	0.5	140	22	23	166	7.2	10.1	3.2	3.5	Nil	Nil	nil	1-2	1-2
2	C81264	39	F	7200	53	42	5	0	2.5	96	118	16	0.5	145	31	33	180	6.5	10.3	3.2	6.4	Nil	Nil	nil	2-4	2-4
3	C75674	55	F	8000	65	26	9	0	4.1	92	112	14	0.4	150	14	15	175	5.5	10	3.5	2.9	Nil	Nil	nil	2-4	2-4
4	C83901	25	F	6300	55	42	3	0	3.8	93	104	14	0.4	119	15	16	130	4.6	10.5	3.1	2.9	Nil	Nil	nil	2-4	2-4
5	C82169	45	M	9400	59	36	4	1	2.9	84	120	15	0.4	200	20	22	152	7.1	11.9	3.2	5	Nil	Nil	nil	2-4	2-4
6	C69781	36	F	5200	51	41	8	0	2.9	94	101	14	0.4	105	14	17	149	5.1	10.9	2.9	3	Nil	Nil	nil	2-4	2-4
7	C91254	39	F	4500	58	38	4	0	2	95	108	18	0.5	115	30	32	196	6.5	10.8	3	3	Nil	Nil	nil	1-2	1-2
8	C92794	33	F	9300	49	34	16	1	2.2	101	116	16	0.5	114	30	32	189	6.6	10.1	2.8	4	Nil	Nil	nil	1-2	1-2
9	C91728	27	F	8900	70	26	4	0	4.1	98	116	23	0.7	143	11	12	155	6.6	11	3.4	3.5	Nil	Nil	nil	4-8	3-6
10	C92876	40	F	5400	40	55	5	0	2.6	101	120	17	0.4	167	12	11	145	6.5	10.1	2.7	3.4	Nil	Nil	nil	1-2	2-3
11	C92882	51	F	6100	63	35	2	0	4.7	102	136	14	0.4	178	14	16	154	6.7	10.7	2.9	3.7	Nil	Nil	nil	2-3	2-3
12	C82212	28	M	9600	62	35	3	0	2.8	94	104	18	0.5	189	25	27	166	7.5	11.5	3	7.1	Nil	Nil	nil	2-4	2-4
13	C93377	37	F	5500	42	55	3	0	2.8	97	120	14	0.4	150	19	21	170	7	10.8	2.9	3	Nil	Nil	nil	2-3	2-3
14	C91376	37	F	6000	50	46	4	0	3.8	87	114	15	0.6	137	19	21	140	7	10	2.8	4	Nil	Nil	nil	1-2	1-2
15	C94453	49	F	4800	56	39	5	0	2.7	91	140	14	0.5	161	19	21	175	6	10.4	2.9	4.3	Nil	Nil	nil	2-4	2-4
16	C89619	38	F	7700	60	38	2	0	2.7	105	135	14	0.4	189	16	17	195	6	10	2.8	4	Nil	Nil	nil	1-2	2-3
17	C94419	46	F	7200	53	44	3	0	4.1	92	114	14	0.4	180	20	22	183	5.5	10.6	2.8	4.3	Nil	nil	nil	1-2	1-2
18	4248	48	F	5900	68	28	4	0	4.5	83	129	14	0.5	182	12	15	160	6.2	10.5	3.3	5	Nil	nil	nil	1-2	1-2
19	D000707	47	M	5400	65	30	5	0	2.7	84	104	21	0.9	163	17	19	166	5.9	10.4	3	3.7	Nil	nil	nil	2-4	2-4
20	C90994	28	F	7700	55	39	6	0	2.9	96	132	14	0.4	139	11	12	153	6.1	10.6	3	3	Nil	nil	nil	2-4	2-4

IMPROVEMENT IN PROGNOSIS OF SYMPTOMS OF PANDU

S.NO	OP/IP NO	AGE	SEX	PALLOR		BREATHLESSNESS		TIREDNESS		GIDDINESS		ANOREXIA		PICA		PALPITATIONS	
				BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT	BT	AT
1	C81998	52	F	+	+	+	_	+	_	_	_	+	_	+	_	+	_
2	C81264	39	F	+	+	+	_	+	+	+	_	+	+	+	+	+	+
3	C75674	55	F	+	+	+	_	+	_	_	_	+	_	_	_	_	_
4	C83901	25	F	+	+	+	_	+	_	+	_	+	_	+	_	+	_
5	C82169	45	M	+	+	_	_	_	_	_	_	_	_	_	_	+	_
6	C69781	36	F	+	+	+	+	+	_	+	+	+	_	+	+	+	_
7	C91254	39	F	+	_	+	_	+	_	_	_	+	_	_	_	_	_
8	C92794	33	F	+	_	+	_	+	_	_	_	+	_	+	_	+	_
9	C91728	27	F	+	+	+	_	+	_	+	_	+	+	+	+	+	_
10	C92876	40	F	+	+	+	+	+	_	+	_	+	_	+	_	+	_
11	C92882	51	F	+	+	+	+	+	+	+	+	+	_	+	_	+	+
12	C82212	28	M	+	+	+	_	+	_	_	_	+	_	_	_	+	_
13	C93377	37	F	+	+	+	+	+	+	+	_	+	_	+	+	+	+
14	C91376	37	F	+	+	+	_	+	_	+	_	+	_	+	_	+	_
15	C94453	49	F	+	+	_	_	+	_	+	_	+	_	_	_	_	_
16	C89619	38	F	+	+	+	_	_	_	_	_	_	_	_	_	_	_
17	C94419	46	F	+	+	+	_	+	_	_	_	+	_	+	_	+	_
18	4248	48	F	+	+	+	_	+	_	+	_	+	_	+	_	+	_
19	D000707	47	M	+	+	_	_	+	_	+	_	+	_	_	_	+	_
20	C90994	28	F	+	+	+	_	+	_	+	_	+	+	+	_	+	_

TABLES FOR TRAIL DRUG-2 SINGATHI CHOORNAM

TRIAL DRUG II, TABLE 1: QUALITATIVE ANALYSIS:

S.NO	PARAMETERS	RESULTS
1.	Phosphate	Absent
2.	Sulphate	Absent
3.	Magnesium	Present
4.	Iron	Present
5	Aminoacids	Present
6.	Starch	Absent
7.	Flavonoids	Absent
8.	Proteins	Absent
9.	Tannic acid	Present
10.	Glycosides	Absent

TRIAL DRUG II, TABLE 2: PHYSICAL PROPERTIES

S.NO	Characteristic test	Results
1.	Ph	5.52
2.	Ash Value	0.85
3.	Water soluble ash	0.11
4.	Acid insoluble ash	0.13

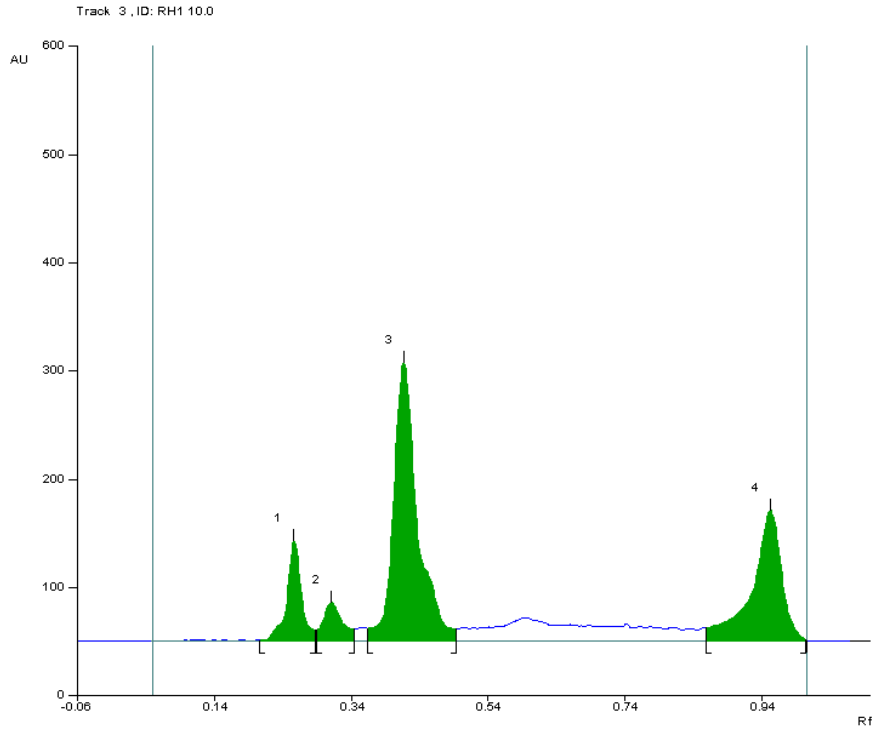
TRIAL DRUG II, TABLE 3: Preliminary acid, basic radicals screening of Singathi Chooranam

S.No.	Constituents	SC
1.	magnesium	+
2.	Iron (Ferric)	+
3.	Iron (Ferrous)	+
4.	sulphate	_
5.	Sodium	+
6.	Starch	_
7.	Phosphate	_

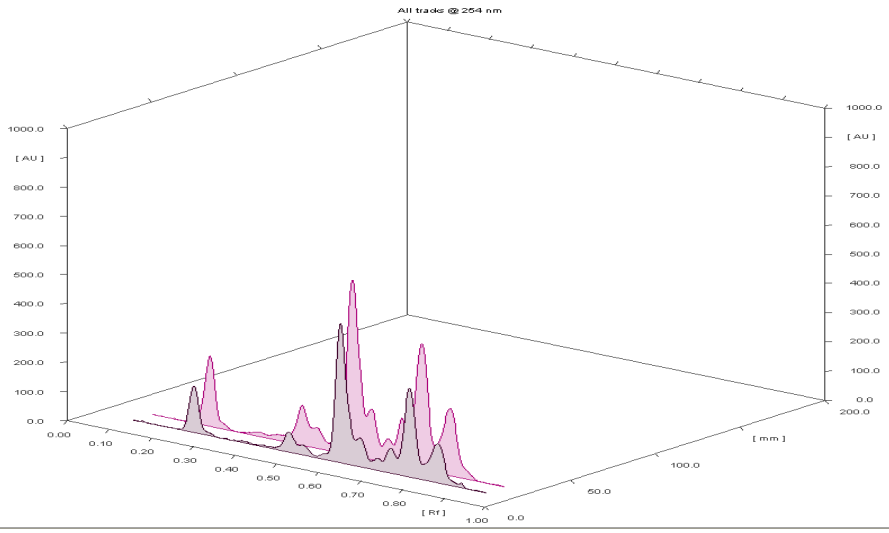
TRIAL DRUG II, TABLE 4: METAL CONTENT:

SAMPLE NAME	Fe (ppm)	Zn (ppm)	K (ppm)	Na (ppm)	Ca (ppm)	Mg (ppm)
SINGATHI CHOORANAM	1.658	_	_	NA	_	_

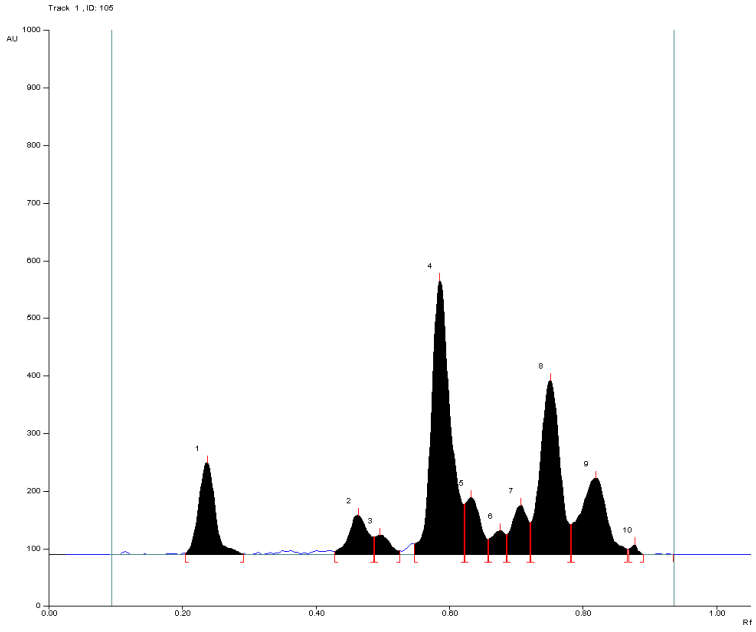
TRIAL DRUG II, GRAPH 1: FINGER PRINTING



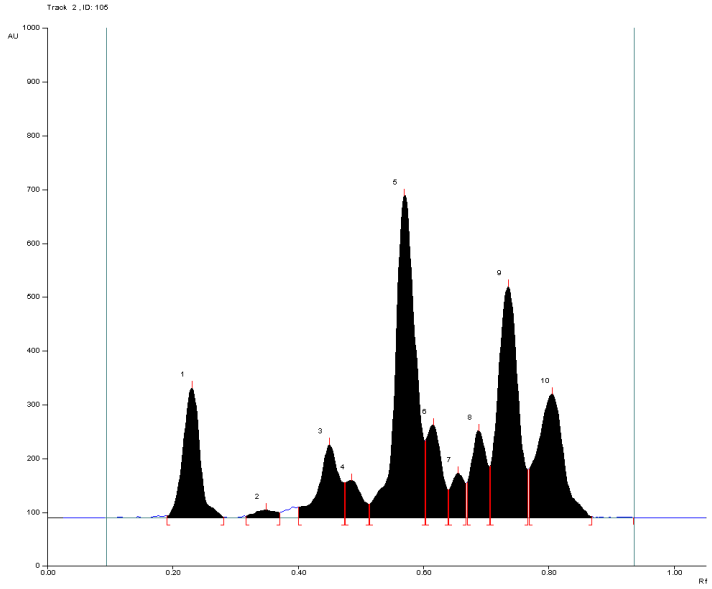
254nm



254nm at 3D Display (*No: 105 -01*)

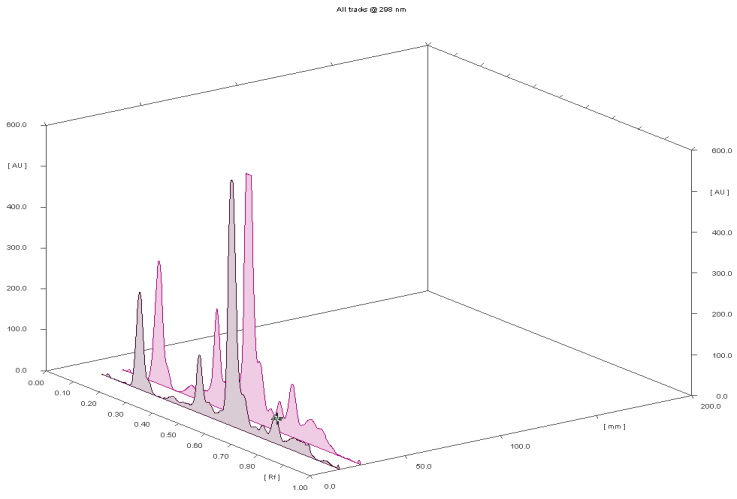


5µl at 254nm (*No: 105 -02*)

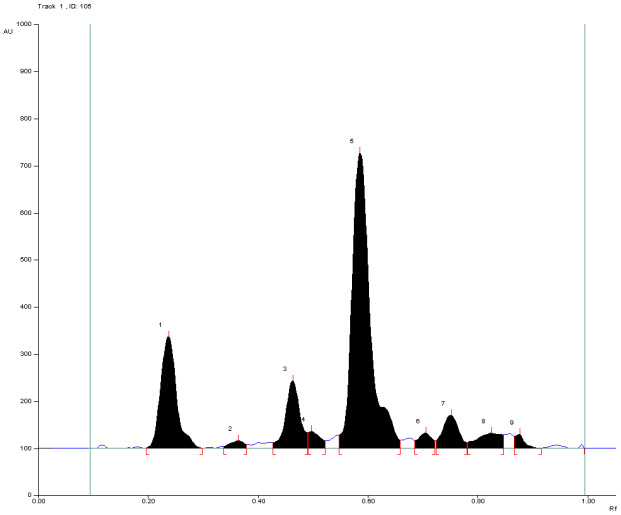


10 µl at 254nm (*No: 105 -03*)

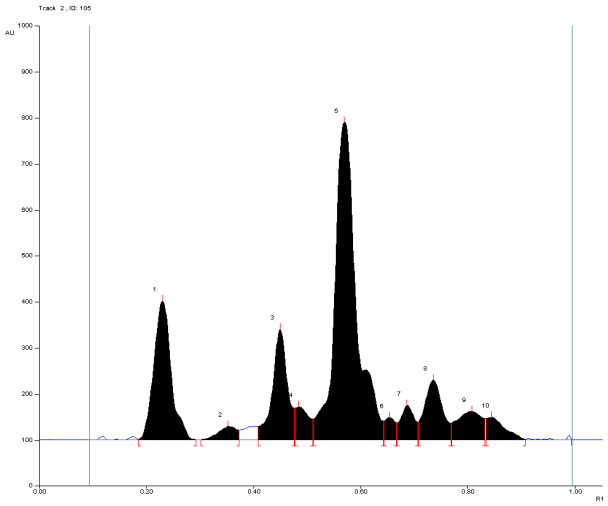
298nm



298 nm at 3D Display (*No: 105 -04*)

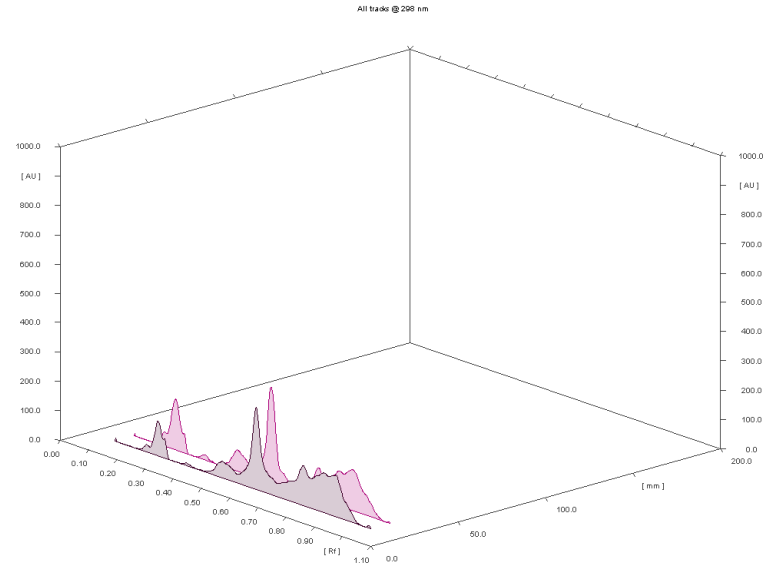


5µl at 298nm (*No: 105 -05*)

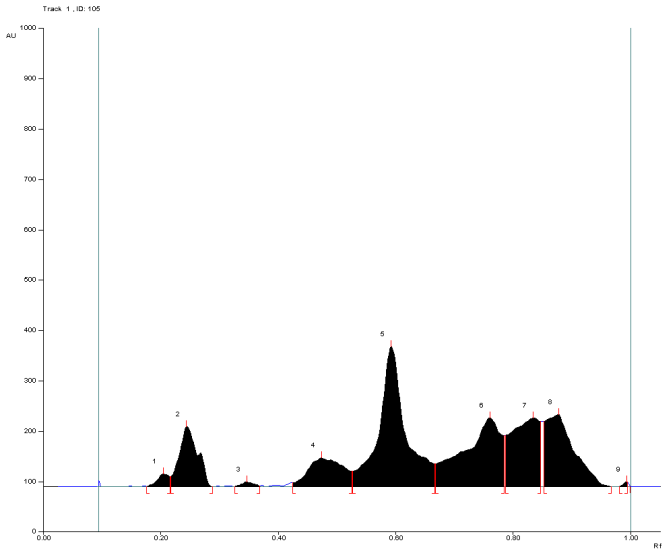


10µl at 298nm (*No: 105 -06*)

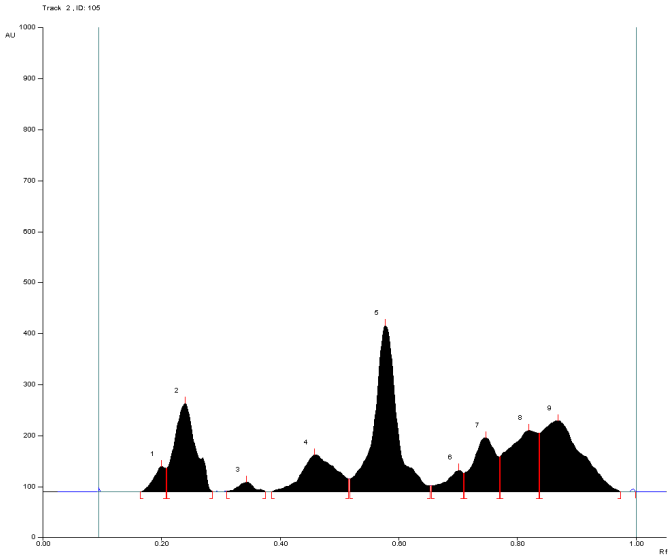
Derivatisation (298nm)



298 nm at 3D Display_ (No: *105 -07*)

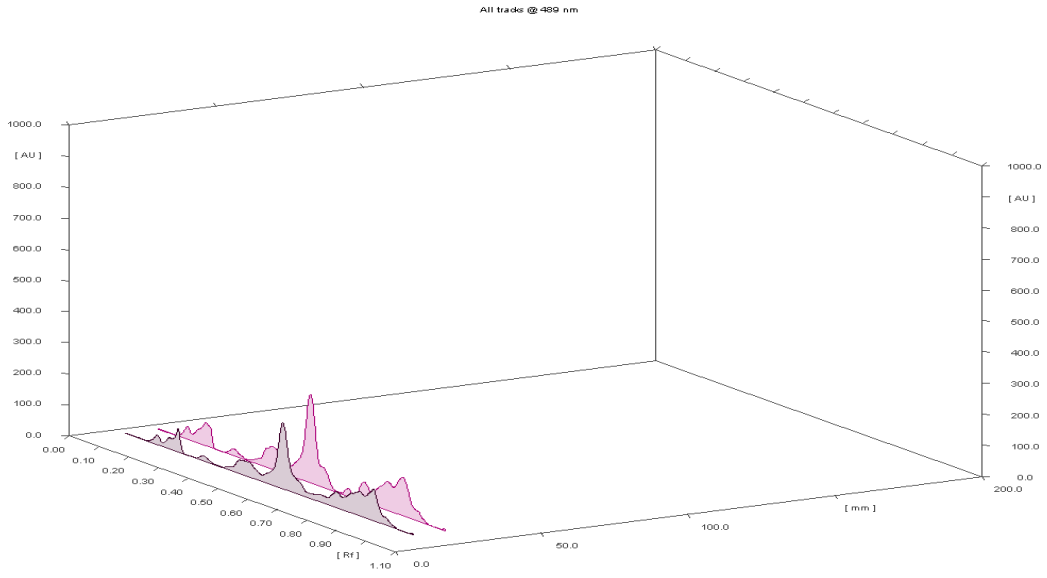


Derivatisation 5µl at 298nm (*No: 105 -08***)**

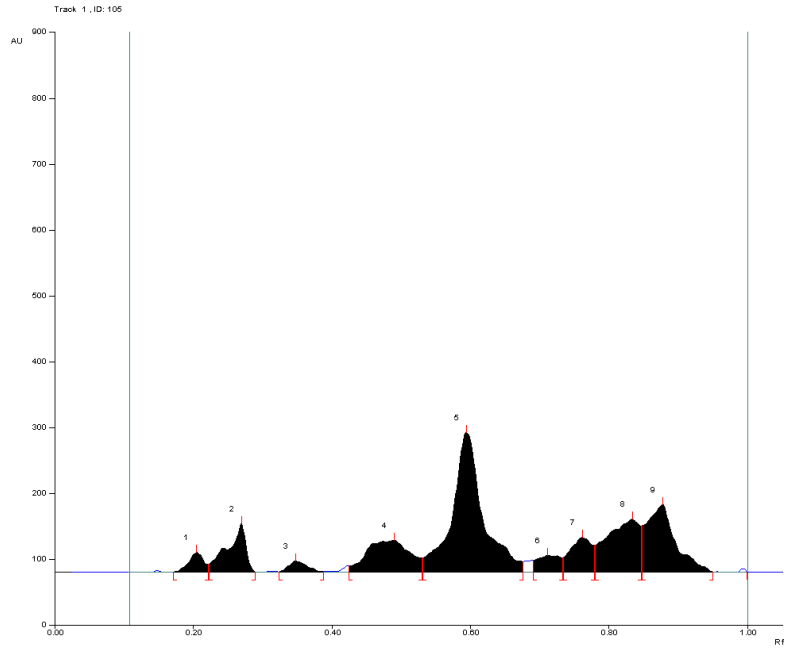


Derivatisation 10µl at 298nm (*No: 105 -09***)**

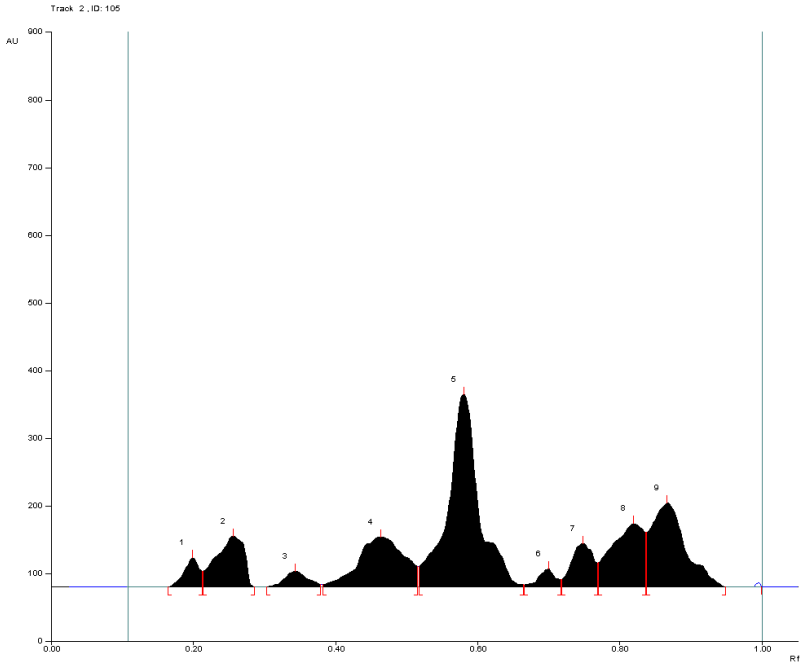
Derivatisation (489nm)



489 nm at 3D Display (*No: 105 -10*)

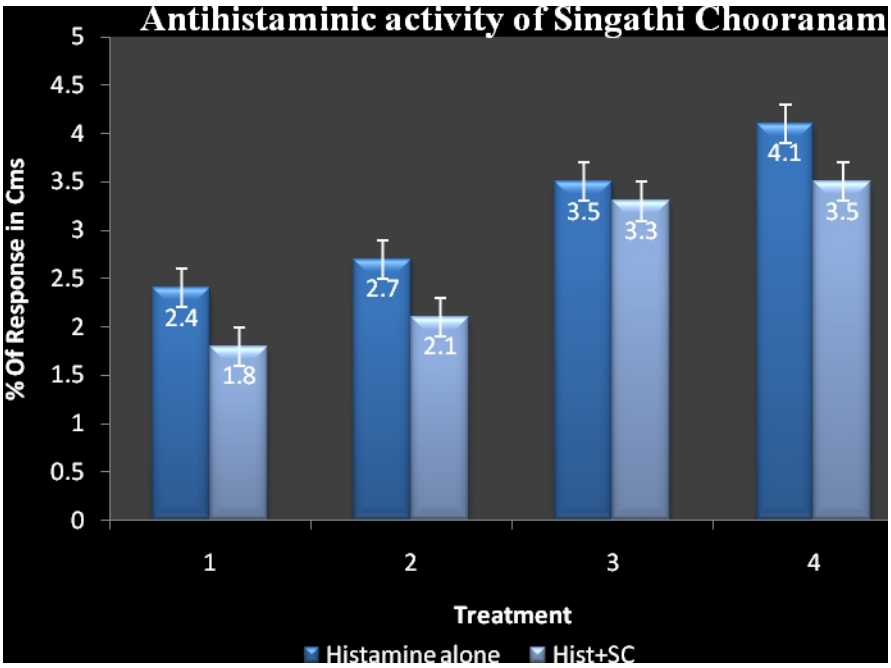


Derivatisation 5µl at 489nm (*No: 105 -11*)

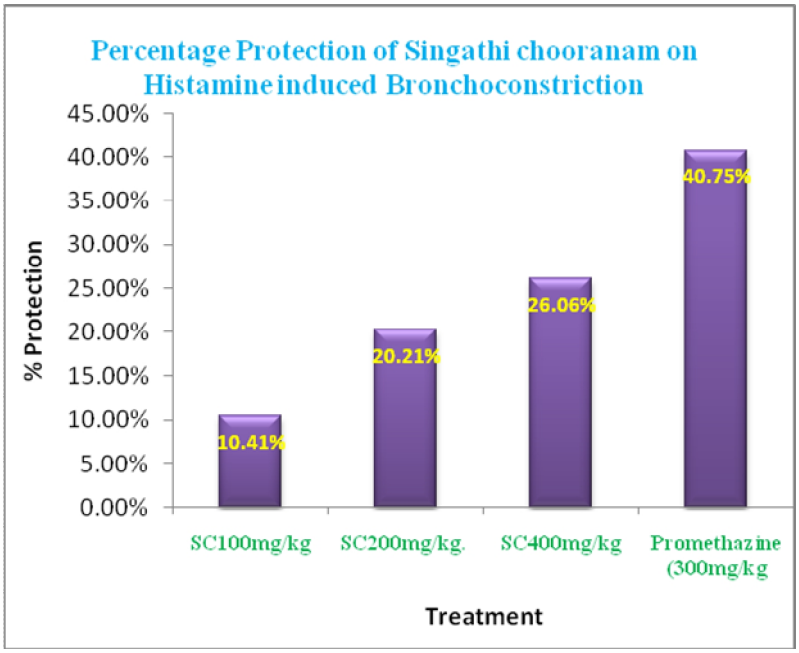


Derivatisation 10µl at 489nm (*No: 107 -12*)

TRIAL DRUG II, CHART 1



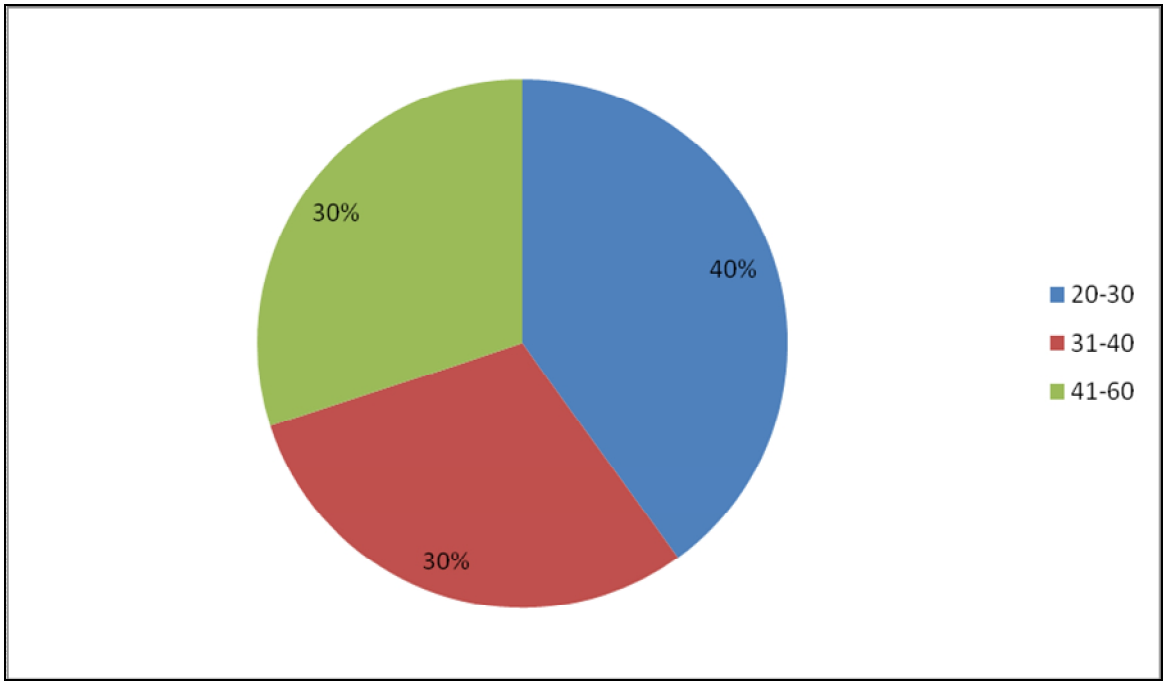
TRIAL DRUG II, CHART 2



CLINICAL OBSERVATION

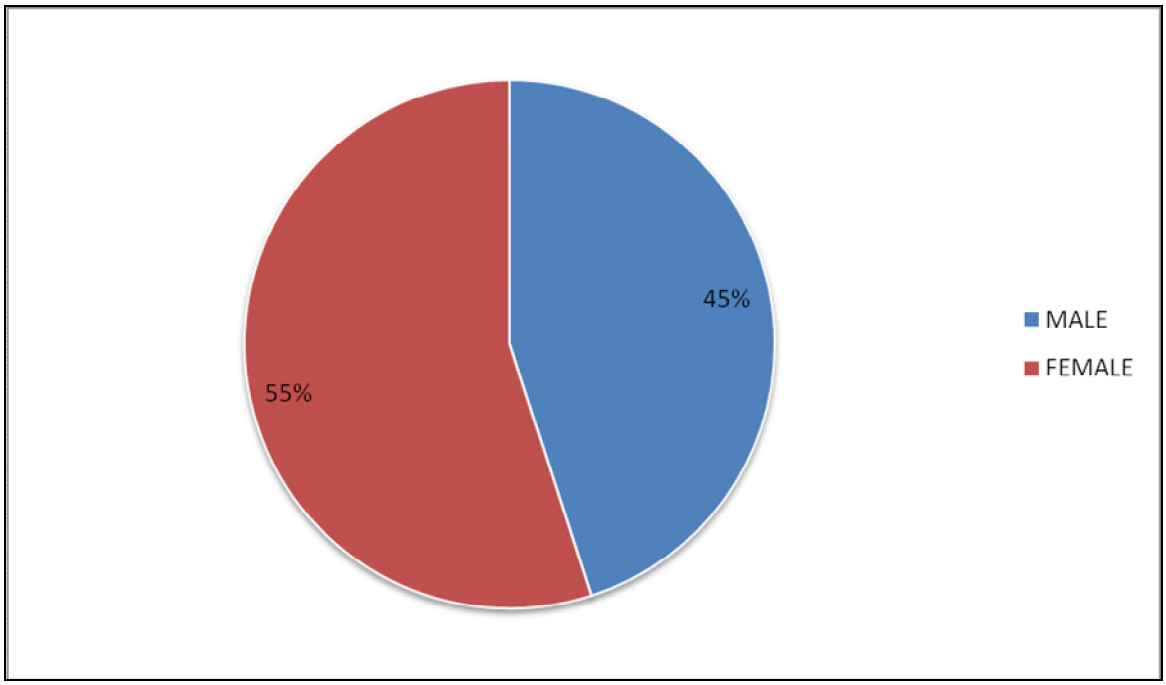
TRIAL DRUG II, TABLE 15:AGE DISTRIBUTION

S.NO	AGE(YEARS)	PERCENTAGE
1.	20-30	(8) 40%
2.	31-40	(6) 30%
3.	41-60	(6) 30%



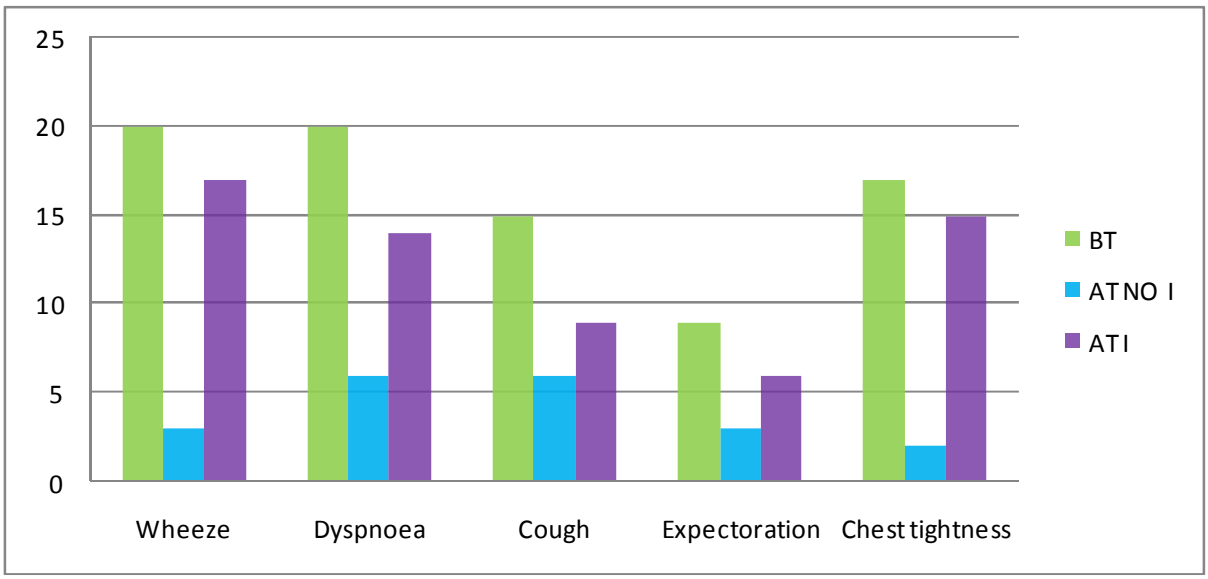
TRIAL DRUG II, TABLE 16: GENDER DISTRIBUTION

S.NO	GENDER	NO OF PATIENTS	PERCENTAGE
1.	MALE	9	45%
2.	FEMALE	11	55%



TRIAL DRUG II, TABLE 17: IMPROVEMENT SHOWING SIGNS AND SYMPTOMS BEFORE AND AFTER TREATMENT OF ERAIPPU PATIENTS.

S.NO	SYMPTOM	NO OF PATIENTS WITH SYMPTOMS			
		BT	AT		Improvement Percentage
			No improvement	Improvement	
1.	Wheeze	20(100%)	3(15%)	17(85%)	85%
2.	Dyspnoea	20(100)%	6(30%)	14(70%)	70%
3.	Cough	15(75%)	6(40%)	9(60%)	60%
4.	Expectoration	9(45%)	3(33.33%)	6(66.67%)	66.67%
5.	Chest tightness	17(85%)	2(11.77%)	15(88.23%)	88.23%



IMPROVEMENT IN PEFR

S.NO	OP/IP NO	AGE	SEX	BT PEFR	AT PEFR
1	C71982	42	M	220	260
2	C84528	47	F	200	240
3	C83020	51	F	150	170
4	C86752	38	F	230	230
5	4042	29	**F**	200	240
6	C87417	35	M	220	290
7	C90579	47	F	220	260
8	C77301	24	F	160	220
9	4086	35	F	170	250
10	C85121	21	M	220	270
11	5043	23	M	220	340
12	4097	36	F	230	280
13	C91984	41	M	250	250
14	C92779	27	F	200	260
15	C92780	37	F	180	180
16	5077	60	M	130	200
17	C57806	39	M	230	300
18	C97429	31	M	250	250
19	C94448	39	F	180	260
20	C74388	51	M	220	320

STATISTICAL ANALYSIS

All collected data were entered into MS Excel software using different columns as variables and rows as patients. SPSS software was used to perform statistical analysis. Basic descriptive statistics include frequency distributions and cross-tabulations were performed. The quantity variables were expressed as Mean ± Standard Deviation and qualitative data as percentage. A probability value of <0.05 was considered to indicate as statistical significance. Paired 't' test was performed for determining the significance between before and after treatment.

Paired t test for Symptoms before and after treatment:

Variable	Obs	Mean	Std.dev	t.value	P value
Before symptoms	20	4.05	0.759	12.990	P<0.0001
After symptoms	20	1.00	0.973		

For Symptoms, the mean and standard deviation before treatment is 4.05±0.759 and after treatment is 1.00±0.973, which is statistically significant(p<0.0001).

Paired t test for AEC before and after treatment:

Variable	Obs	Mean	Std.dev	t.value	P value
Before treatment	20	524.65	169.341	4.283	P<0.0001
After treatment	20	320.35	189.553		

For AEC, the mean and standard deviation before treatment is 524.65±169.341 and after treatment is 320.35±189.553, which is statistically significant(p<0.0001).

Paired t test for PEFR before and after treatment:

Variable	Obs	Mean	Std.dev	t.value	P value
Before treatment	20	204.00	32.831	-6.521	P<0.0001
After treatment	20	253.50	42.212		

For PEFR, the mean and standard deviation before treatment is 204.00±32.831 and after treatment is 253.50±42.212, which is statistically significant(p<0.0001).

TOXICOLOGY TABLES

Trial drug II, Table 5: Dose finding experiment and its behavioural Signs of Toxicity

No	Dose mg/kg	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
1.	**500**	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
2.	**1000**	+	-	-	+	-	+	-	-	-	-	-	-	-	-	-	-	-	-	-	-
3.	**2000**	+	-	-	+	-	+	+	-	-	-	-	-	+	-	-	-	-	+	+	-

1. Alertness 2. Aggressiveness 3. Pile erection 4. Grooming 5. Gripping 6. Touch Response 7. Decreased Motor Activity 8. Tremors 9. Convulsions 10. Muscle Spasm 11. Catatonia 12. Muscle relaxant 13. Hypnosis 14. Analgesia 15.Lacrimation 16. Exophthalmos 17. Diarrhoea 18. Writhing 19. Respiration 20. Mortality

Trial drug II, Table 6. Body wt (g) of rats exposed to *Singathi Chooranam* for 28days.

Dose (mg/kg/day)	Days				
	1	**7**	**14**	**21**	**28**
Control	120.19±4.00	122.10±3.44	125.19±4.34	128.14±5.11	130.04±3.56
100	122.64±4.18	124.75±4.18	127.33±4.12	129.20±5.52	130.14±4.45
200	127.10±4.00	129.50±3.55	124.15±3.88	126.12±4.84	128.16±4.34
400	124.16±3.54	126.44±4.32	128.10±4.10	130.04±3.96	132.12±5.20

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$; Vs Control N=6.

Trial drug II, Table 7. Food intake of rats exposed to *Singathi Chooranam* for 28days.

Dose (mg/kg/day)	Days (gms/rats)				
	1	**7**	**14**	**21**	**28**
Control	42.46±2.88	45.11±2.50	50.74±2.38	50.14±2.62	52.40±3.00
100	40.20±2.51	45.20±2.46	42.23±2.88	50.49±2.24	48.14±3.41
200	41.15±2.17	42.56±2.34	44.50±2.64	45.53±3.00	46.30±3.12
400	45.24±2.22	45.46±2.57	45.22±2.50	45.51±2.89	45.10±3.14

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$; Vs Control N=6.

Trial drug II, Table 8. Water (ml/day) intake of rats exposed to *Singathi Chooranam* for 28days.

Dose (mg/kg/day)	Days(ml/rat)				
	1	**7**	**14**	**21**	**28**
Control	52.20±2.12	52.18±3.04	52.46±3.02	52.14±3.20	51.15±3.00
100	51.11±2.36	52.12±2.88	55.00±3.12	54.22±3.05	55.12±2.41
200	42.26±2.48*	44.10±3.00	42.73±3.10	42.20±2.31	42.10±3.50
400	54.00±3.44	52.17±3.13	51.42±3.00	52.11±2.62	50.52±3.00

Values are mean ± S.E.M. (Dunnet 't' test). *P<0.05; Vs Control N=6.

Trial drug II, Table 9. Hematological parameters after 28days treatment with *Singathi Chooranam*.

Parameter	Control	100 mg/kg	200 mg/kg	400 mg/kg
RBC (millions/cu.mm)	5.01±0.32	5.12±0.35	5.15±0.52	5.44±0.36
Hb (g/dl)	13.47±0.78	13.99±0.86	14.2±1.4	14.00±1.2
PCV (%)	44.18±3.12	44.05±3.10	44.10±2.92	44.30±2.81
WBC(cells/cu.mm)	7882±361.4	8324±312.4	8417±368.1	8542±225.2
Neutrophil (%)	45.10±2.18	42.36 ±2.78	43.00±3.1	42.45±3.0
Lymphocytes (%)	44.25±2.45	43.26±2.44	43.28±2.56	44.34±2.2
Eosinophil's (%)	3.7±0.42	3.2±0.50	3.4±0.41	3.5±0.38
Monocytes (%)	4.2±0.30	4.1±0.31	4.2±0.4	4.1±0.4
Basophils (%)	0±0	1.0±0	0±0	0±0
Platelets (10^5 cells/cu.mm)	1.67±0.07	1.52±0.05	1.75±0.7	1.62±0.5
MCV(Fl)	72.2±1.8	69.2±1.2	73.4±2.0	73.1±2.1
MCHC (pg)	29.5±2.2	28.41±2.4	29.2±2.1	30.00±2.5

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$; Vs Control N=6.

Trial drug II, Table 10. Effect of *Singathi Chooranam* biochemical (LFT, RFT, Lipid Profile) Parameters

Parameter	Control	100 mg/kg	200 mg/kg	400 mg/kg
Glucose (mg/dL)	82.22±4.10	80.02±4.48	79.75±3.88	78.62±4.64
Total Bilirubin (mg/dL)	0.210±0.03	0.212±0.05	0.214±0.05	0.212±0.04
Bilirubin direct (mg/dL)	0.1±0.03	0.1±0.02	0.1±0.03	0.1±0.04
Creatinine (mg/dL)	0.91±0.05	0.93±0.04	0.92±0.04	0.90±0.04
BUN (mg/dL)	17.24±1.48	18.02±1.32	17.58±1.46	17.85±1.39
AST (IU/L)	128.1±5.28	126.4±5.10	125.4±5.22	127.4±5.72
ALT (IU/L)	35.14±3.15	34.48±2.99	35.00±2.46	34.92±2.75
ALP (IU/L)	66.11±4.41	67.10±3.82	66.12±4.30	65.41±4.44
Total cholestrol (mg/dL)	56.10±5.10	57.24±5.52	56.42±5.44	56.19±4.92
Total protein (g/dL)	7.20±0.48	7.48±0.54	7.42±0.70	7.50±0.72
Albumin (g/dL)	2.81±0.07	2.72±0.05	2.70±0.05	2.75±0.05
Urea(mg/dL)	55.00±2.48	54.78±3.36	55.42±2.48	54.44±2.12
Uric acid (mg/dL)	1.7±0.10	1.6±0.12	1.6±0.14	1.47±0.10
Na m.mol	143.66±5.00	142.8±4.12	140.32±4.21	140.41±5.10
K m.mol	20.24±2.56	19.00±1.05	20.14±1.78	20.47±1.86
Cl m.mol	103.11±4.21	104.12±4.10	103.24±4.24	102.12±4.11
HDL(mg/dL)	13.33±1.00	13.14±1.22	13.17±1.34	13.28±1.45
LDL(mg/dL)	43.34±2.21	43.38±3.42	42.14±3.13	42.12±3.20
VLDL(mg/dl)	15.45±2.62	15.90±2.60	15.48±2.35	15.96±2.12
Triglycerides (mg/dl)	86.38±3.44	86.38±2.45	87.42±2.21	88.30±2.58

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$; Vs Control N=6.

Trial drug II, Table-11 Urine Analysis

Parameters	Control	100 mg/kg	200 mg/kg	400 mg/kg
Colour	Yellow	Yellow	Yellow	Yellow
Transparency	Clear	Slightly turbid	Slightly cloudy	Slightly turbid
Specific gravity	1.010	1.010	1.010	1.010
PH	>7.2	>8.0	>8.0	>9.0
Protein	Nil	3+	3+	3+
Glucose	Nil	Nil	Nil	Nil
Bilirubin	-ve	-ve	-ve	-ve
Ketones	-ve	+ve	+ve	+ve
Blood	Absent	Absent	Absent	Absent
Urobilinogen	Normal	Abnormal	Abnormal	Abnormal
Pus cells	0-cells/HPF	1-cell/HPF	2-cells/HPF	1-cell/HPF
RBCs	Nil	Nil	0-1cells/HPF	Nil
Epithelial cells	Nil	1-cell/HPF	Nil	1-cell/HPF
Crystals	Nil	Nil	Nil	Nil
Casts	Nil	Nil	Nil	Nil

Trial drug II, Table 12. Effect of *Singathi Chooranam* on organ weight

Dose (mg/kg)	Control	100 mg/kg	200 mg/kg	400 mg/kg
Heart(g)	0.68±0.08	0.68±0.10	0.64±0.08	0.68±0.04
Liver(g)	4.63±0.40	4.70±0.72	4.83±0.60	4.72±0.50
Lung(g)	0.69±0.04	0.70±0.05	0.71±0.05	0.70±0.4
Spleen(g)	0.69±0.04	0.68±0.04	0.70±0.05	0.69±0.05
Kidney(g)	1.04±0.3	1.02±0.04	1.52±0.6	1.44±0.04
Testis(g)	0.91±0.03	0.92±0.05	0.91±0.04	0.91±0.03
Ovary(g)	0.04±0.01	0.04±0.02	0.04±0.02	0.04±0.02
Brain	0.70±0.05	0.68±0.04	0.67±0.04	0.68±0.05
Pancreas	1.42±0.04	1.44±0.06	1.48±0.12	1.50±0.10
Uterus	0.78±0.04	0.84±0.07	0.80±0.04	0.80±0.05

Values are mean ± S.E.M. (Dunnet 't' test). $^{ns}P>0.05$; Vs Control N=6.

PHARMACOLOGICAL TABLES

Trial drug II, Table-13: Effect of Singathi Chooranam on isolated Guinea pig ileum preparation

S. No	Dose of Histamine (µg/ml)	Percent of maximum response	
		Histamine alone	Histamine+Singathi Chooranam (1mg/ml)
1	10	2.4±0.7	1.8±0.5
2	20	2.7±0.47	2.1±0.6*
3	40	3.5±0.12	3.3±0.6*
4	80	4.1±0.58	3.5±0.5**

Values are expressed in mean ± SEM, *p< 0.05: **P<0.01 compared with histamine alone induced contraction (41mm as 100%); n=3.

Trial drug II, Table-14: Bronchodilator effect of *Singathi chooranam* on Histamine induced Bronchoconstriction.

Treatment	Pre-Treatment Exposition in seconds	Post-Treatment Exposition in seconds	Percentage Protection
Singathi Chooranam 100mg/kg. p.o.	90.54 ±4.50	101.07±4.26	10.41%
Singathi Chooranam 200mg/kg. p.o.	98.22±3.22	123.11±4.71**	20.21%
Singathi Chooranam 400mg/kg. p.o.	94.28±3.46	127.52±4.66**	26.06%
Promethazine (300mg/kg, p.o)	102.12±4.45	172.38±4.41**	40.75%

N=6; Values are expressed as mean ± SEM; *Significant between pre and post treatment time (Student's –'t') **P<0.01.

ERAIPPU - BEFORE INVESTIGATIONS

S.NO	OP/IP NO	AGE	SEX	HB [g/dl]	TC [Cu. Mm]	DC				TRBC [million/ Cu.mm]	PLT Lakhs/ Cu.mm	BLOOD SUGAR [mg/dl]		UREA [mg/ dl]	CREAT [mg/ dl]	CHOL [mg/ dl]	LFT				CA [mg/ dl]	PHOS [mg/ dl]	U.A [mg/ dl]	URINE					AFB
						P%	L%	E%	M%			F	PP				SGOT Iu/L	SGPT Iu/L	ALP Iu/ L	TP mg/dl				ALB	SUGAR		DEP		
																									F	PP	PUS	EPI	
1	C71982	42	M	15.8	10400	63	30	7	0	5.3	3.6	79	120	17	0.6	161	23	24	160	7.1	10.1	3.1	5.4	nil	nil	nil	1-2	1-2	neg
2	C84528	47	F	12.9	8200	60	23	16	1	4.2	3.7	91	108	17	0.5	224	18	19	138	6.5	10	3.5	6	nil	nil	nil	2-3	2-3	neg
3	C83020	51	F	12.1	6500	60	33	7	0	4.2	2.8	98	112	37	0.8	221	20	22	140	7	9.6	3.4	5.5	nil	nil	nil	2-4	2-4	neg
4	C86752	38	F	12.3	7800	58	30	12	0	4.2	1.8	103	125	17	0.5	142	14	15	156	7.6	9	3.2	5.3	nil	nil	nil	2-4	1-2	neg
5	4042	29	F	13.2	11400	74	22	4	0	4.5	2.6	73	89	22	0.6	145	22	27	168	7	10.2	2.6	4.4	nil	nil	nil	3-4	3-4	neg
6	C87417	35	M	15.1	8000	66	20	14	0	4.9	2.1	101	129	14	0.4	93	22	23	150	7.2	10.8	3.2	6.2	nil	nil	nil	2-4	2-4	neg
7	C90579	47	F	13.2	9400	65	30	6	0	4.7	3.7	107	119	15	0.4	162	26	27	147	6.9	10.5	3.2	5.6	nil	nil	nil	2-4	2-4	neg
8	C77301	24	F	14	12800	56	34	10	0	5.3	2.9	92	106	22	0.6	167	20	21	130	7.3	10.7	3	4.9	nil	nil	nil	2-4	2-4	neg
9	4086	35	F	12.1	9400	64	30	6	0	4.2	2.3	81	105	24	0.7	166	15	16	140	7.5	10.5	3.2	6	nil	nil	nil	1-2	1-2	neg
10	C85121	21	M	15.9	8500	70	20	10	0	5.4	2.9	94	114	14	0.4	91	20	21	176	6.6	11	3.6	3.2	nil	nil	nil	1-2	1-2	neg
11	5043	23	M	15.9	4800	56	37	7	0	5.6	2.4	75	89	18	0.5	83	27	24	166	5.7	11.6	3.4	7.6	nil	nil	nil	2-3	2-3	neg
12	4097	36	F	13	9400	52	43	5	0	4.4	2.3	73	95	16	0.5	108	11	10	135	6.9	10.8	2.9	5	nil	nil	nil	1-2	1-2	neg
13	C91984	41	M	16.3	6100	34	57	9	0	5.2	2	105	114	32	0.8	197	14	16	150	7.3	10.6	3.1	5.1	nil	nil	nil	3-4	2-3	neg
14	C92779	27	F	13	7200	70	20	9	1	4.3	3	89	117	24	0.6	151	11	12	175	5.7	10.1	2.9	3	nil	nil	nil	1-2	2-4	neg
15	C92780	37	F	13.4	8600	52	40	7	1	4.4	3.2	103	131	30	0.7	106	12	15	136	5.6	10.6	3	3.2	nil	nil	nil	2-4	2-4	neg
16	5077	60	M	12.4	6600	69	22	9	0	4.1	2.7	89	118	19	0.5	127	20	22	184	5.7	11	3.1	3.4	nil	nil	nil	1-2	1-2	neg
17	C57806	39	M	13.2	8800	59	30	10	1	6.2	2.5	90	123	23	0.6	163	20	22	179	6.5	10	2.9	5	nil	nil	nil	4-5	3-4	neg
18	C97429	31	M	16.8	6900	57	30	13	0	5.2	1.9	101	117	20	0.6	146	15	17	176	6.6	10	3.1	4.4	nil	nil	nil	2-3	2-3	neg
	C94448	39	F	12.3	4700	58	31	11	0	4.1	2.5	84	101	17	0.5	142	12	14	160	5.7	10.2	2.9	3	nil	nil	nil	1-2	1-2	neg
20	C74388	51	M	13.6	7900	54	35	10	2	4.8	2.3	83	108	18	0.5	171	30	33	148	7.1	10.6	3.5	5	Nil	nil	nil	1-2	2-3	neg

ERAIPPU -AFTER INVESTIGATIONS

S.NO	OP/IP NO	AGE	SEX	HB [g/dl]	TC [cu.mm]	DC				PLT [million/ Cu. mm]	TRBC Lakhs/ Cu.mm	BLOOD SUGAR [mg/dl]		URE [mg/dl]	CREA [mg/dl]	CHO [mg/dl]	LFT				CA [mg/dl]	PHO [mg/dl]	U.A[mg/dl]	URINE					AFB
						P %	L %	E %	M %			FBS	PPBS				SGOT [Iu/L]	SGPT Iu/L]	ALP Iu/L]	TP [mg/dl]				ALB	SUGAR		DEP		
																									F	PP	PUS	EPI	
1	C71982	42	M	15.2	10100	65	30	5	0	5.2	3.5	81	100	15	0.5	153	35	25	194	6.1	12	3.5	6.8	nil	nil	nil	2-3	2-3	neg
2	C84528	47	F	13	7900	59	31	10	0	4.2	3.9	100	112	15	0.4	220	14	15	141	7.5	10	3	3.9	nil	nil	nil	2-4	3-5	neg
3	C83020	51	F	12.1	8600	57	39	4	0	4.2	2.7	100	120	21	0.6	181	29	30	166	6.6	10	3.2	3.7	nil	nil	nil	1-2	1-2	Neg
4	C86752	38	F	13	8100	56	30	12	2	4.5	3.7	102	120	19	0.5	159	12	14	140	5.8	11	3.4	4.3	nil	nil	nil	2-3	2-3	Neg
5	4042	29	**F**	13.2	10400	68	29	3	0	4.5	2.4	75	99	14	0.5	105	12	14	166	5.7	11	3.1	5	nil	nil	nil	1-2	1-2	Neg
6	C87417	35	M	15	7600	61	34	5	0	4.5	2.1	81	100	19	0.5	101	32	39	197	6.6	11	3.1	5.1	nil	nil	nil	2-4	2-4	Neg
7	C90579	47	F	14	8600	68	29	3	3	5.1	3.1	95	107	15	0.4	130	18	21	142	5.3	10	3.2	3	nil	nil	nil	1-2	2-3	Neg
8	C77301	24	F	14	10900	63	30	6	1	5.2	3.2	86	100	23	0.7	146	16	18	136	7.4	11	3.1	3.1	nil	nil	nil	2-3	1-2	Neg
9	4086	35	F	12.5	10500	64	32	4	0	4.4	3.8	82	107	19	0.6	152	21	22	160	5.6	10	2.9	3.5	nil	nil	nil	1-2	2-3	Neg
10	C85121	21	M	15.6	7200	67	27	5	1	5.3	3	93	109	14	0.4	102	18	20	180	6.6	11	3.2	5.5	nil	nil	nil	3-4	2-3	Neg
11	5043	23	M	16.8	9000	68	34	8	0	5.9	2.8	88	96	18	0.5	83	18	20	174	6.5	11	2.8	4.6	nil	nil	nil	1-2	2-3	Neg
12	4097	36	F	13.5	7600	61	34	3	2	4.5	2.1	81	100	19	0.5	101	32	39	197	6.6	11	3.1	5.1	nil	nil	nil	2-4	2-4	Neg
13	C91984	41	M	16.1	5800	66	24	10	0	5.1	1.7	88	106	26	0.7	253	20	21	179	7	11	3.1	4.1	nil	nil	nil	1-2	2-3	Neg
14	C92779	27	F	11.9	7300	59	35	5	1	4.3	3	92	120	17	0.6	199	13	14	141	6.7	11	3	3.1	nil	nil	nil	4-8	4-8	Neg
15	C92780	37	F	11.5	9300	63	30	7	0	4.1	2.8	102	126	14	0.5	126	16	17	139	6.5	11	3.2	3	nil	nil	nil	3-6	4-8	Neg
16	5077	60	M	12.6	4900	59	36	5	0	4.2	2.7	100	118	30	0.8	104	27	28	172	6	11	3.2	3	nil	nil	nil	3-6	3-6	Neg
17	C57806	39	M	13.6	7700	62	33	5	0	5.9	2.8	94	118	18	0.5	160	20	22	192	6	10	2.8	4.7	nil	nil	nil	2-4	2-4	Neg
18	C97429	31	M	15	7200	60	27	13	0	5.2	1.9	105	124	18	0.6	152	20	22	175	6.1	10	3.1	4.5	nil	nil	nil	2-4	2-4	Neg
19	C94448	39	F	12.4	7700	64	32	4	0	4.4	2.4	87	101	26	0.7	173	15	17	173	5.7	9.9	2.8	5	nil	nil	nil	2-3	2-3	Neg
20	C74388	51	M	14.3	8000	61	33	6	0	5	2.4	98	116	14	0.4	163	31	33	215	7.5	9.9	2.9	5.5	nil	nil	nil	2-4	2-4	Neg

SPECIFIC INVESTIGATIONS FOR ERAIPPU

S.NO	OP/IP NO	AGE	SEX	E[%]		½ Hr ESR [mm]		1 Hr ESR [mm]		AEC [cellc/cu.mm]		PEFR	
				BT	AT	BT	AT	BT	AT	BT	AT	BT	AT
1	C71982	42	M	7	5	8	2	14	8	480	260	220	260
2	C84528	47	F	16	10	8	4	18	8	670	480	200	240
3	C83020	51	F	7	4	14	2	22	8	463	370	150	170
4	C86752	38	F	12	12	12	10	26	20	600	600	230	230
5	4042	29	**F**	4	3	4	8	8	12	650	500	200	240
6	C87417	35	M	14	5	6	2	18	4	411	256	220	290
7	C90579	47	F	6	3	12	4	26	12	333	111	220	260
8	C77301	24	F	10	6	4	2	8	10	866	166	160	220
9	4086	35	F	6	4	8	6	26	8	644	56	170	250
10	C85121	21	M	10	5	12	2	16	4	633	88	220	270
11	5043	23	M	7	8	2	2	4	4	277	280	220	340
12	4097	36	F	5	3	2	2	8	4	644	178	230	280
13	C91984	41	M	9	10	12	12	16	16	536	580	250	250
14	C92779	27	F	9	5	10	6	22	10	578	460	200	260
15	C92780	37	F	7	7	8	4	16	10	400	400	180	180
16	5077	60	M	9	5	8	4	28	14	122	64	130	200
17	C57806	39	M	10	5	14	4	22	10	430	200	230	300
18	C97429	31	M	13	13	12	12	20	22	696	706	250	250
19	C94448	39	F	11	4	16	4	24	8	600	346	180	260
20	C74388	25	M	10	6	10	4	18	8	460	306	220	320

IMPROVEMENT IN PROGNOSIS OF SYMPTOMS OF ERAIPPU

S.NO	OP/IP NO	AGE	SEX	WHEEZE		DYSPNOEA		COUGH		EXPECTORATION		CHEST TIGHTNESS	
				BT	AT	BT	AT	BT	AT	BT	AT	BT	AT
1	C71982	42	M	+	_	+	+	+	_	_	_	+	_
2	C84528	47	F	+	_	+	+	+	_	+	_	+	+
3	C83020	51	F	+	_	+	_	_	_	_	_	+	_
4	C86752	38	F	+	_	+	_	+	+	+	_	_	_
5	4042	29	**F**	+	+	+	_	+	+	+	+	+	_
6	C87417	35	M	+	_	+	+	+	+	_	_	+	_
7	C90579	47	F	+	_	+	+	_	_	_	_	+	_
8	C77301	24	F	+	_	+	_	+	_	_	_	+	_
9	4086	35	F	+	_	+	_	+	+	_	_	+	_
10	C85121	21	M	+	_	+	_	+	_	_	_	+	_
11	5043	23	M	+	_	+	_	+	+	+	_	+	_
12	4097	36	F	+	_	+	_	+	_	+	+	+	_
13	C91984	41	M	+	_	+	_	+	+	+	+	_	_
14	C92779	27	F	+	_	+	_	+	_	_	_	+	_
15	C92780	37	F	+	+	+	+	_	_	_	_	+	+
16	5077	60	M	+	_	+	+	+	_	+	_	+	_
17	C57806	39	M	+	_	+	_	+	_	+	_	_	_
18	C97429	31	M	+	+	+	_	+	_	+	_	+	_
19	C94448	39	F	+	_	+	_	_	_	_	_	+	_
20	C74388	51	M	+	_	+	_	_	_	_	_	+	_

BIBILIOGRAPHY

1. http://health.nytimes.com/health/guides/disease.
2. Medical Journal Armed Forces, India,Vol.68,issue 4,Oct 2012.
3. http://www.who.int/vmnis/anaemia/prevalence/en/.
4. Indian Journal Medical Research 130, November 2009.
5. Indian Journal of Haematology and Blood Transfusion,2012.
6. Journal Of Siddha,Vol.2,Jan-June 2009, issue.I.
7. Dr.M.Shanmugavelu,H.P.I.M., Noi Naadal Noi Muthal Naadal thirattu, Part II,2003, Department of Indian Medicine and Homeopathy. 207, 149.
8. Murugesa Mudaliar,Gunapadam Mooligai Vaguppu, Part I, Dept., of Indian Medicine and Homeopathy,7th Edition,2003,808-809.
9. Saraku Suthi Sei Muraigal, Department of Indian Medicine and Homeopathy, Chennai-106, 1st Edition, 2008, Dr.Aanaivaari Ananthan,Ph.D. 4, 6, 7, 13.
10. Kannusamy Parambarai Vaithiyam, B.Rathina nayagar and Sons, Chennai-79, 2006, 155,215,116,243,169,195,239,173,133,107,244,238,330.
11. Dr.A.K.Nadkarni, Indian Materia Medica, Volume I, Ramdas Bhatkal for Popular Prakashan Pvt. Ltd, Mumbai.2005, 170.
12. USDA(United States of Department of Agriculture) Plants Database.
13. Kuppusamy Mudhaliyar,H.P.I.M., Siddha Marurhuvam Pothu, Department of Indian Medicine and Homeopathy,6th Edition, 2004,345-350, 241-245.
14. KV Krishna Das,Textbook of Medicine, 5th Edition,Jaypee Brothers Medical Publishers (P) Ltd,2008,984, 989, 990.
15. R Alagappa, Manual of Practical Medicine,Fourth edition,Jaypee Brothers Medical Publishers (P) Ltd, 2011,347.
16. P.C.Das,Textbook of Medicine,4th Edition,Current Book Inernational, Calcutta,2007,270-271.
17. Thomas M.Delvin, Text Book of Biochemistry with Clinical correlations,Kaye pace-2011,1086.
18. http://www.medicinenet.com
19. Dr.Uththamarayan,H.P.I.M., Thotra Kirama Araichiyum Siddha Maruthuva Varalarum, Department of Indian Medicine and Homeopathy,3rd Edition,2003, 345-350.
20. http://www.thoracomedicine.org/article,2008.

21. D Behera Foreword VK Vijayan,Bronchial Asthma 2nd Edition,2005,Jaypee Brothers Medical Publishers (P) Ltd.
22. http://en.wikipedia.org/wiki/asthma.
23. Kumar and Clark, Clinical Medicine, 5th Edition, 2002, Parveen kumar, Michael clark, WB Saunders, 874.
24. Indian Journal Medical Research, Aug 2011.
25. Harrison's Principles Of Internal Medicine, Volume II, 16th Edition, 2005, Mc Graw Hill, Medical Publishing Division, 1508.
26. http://times of India.indiatimes.com/topic/asthma, 2012.
27. http://en.wikipedia.org/wiki/siddha-medicine.
28. Journal Of National Remedies, Vol.3, issue 2, June 2003.
29. Agathiyar Mani ennum Vaithiya Chinthamani Venba 4000, 1st Edition, 1996, Dr.S.Prema, M.D(S), Thamarai Noolagam, Chennai 26, 183, 252, 254, 269, 365.
30. C.Kannusami pillai, Sigicha Rathna Deepam ennum Vaithiya nool, 1st Edition, 2007, B.Rathina nayagar and Sons, Chennai-79, 28, 29, 30, 35.
31. Dr.R.Thiyagarajan, Gunapadam Thathu Jeeva Vaguppu,Part 2 & 3, 4th Edition, 2004, Department of Indian Medicine and Homeopathy, Chennai-106, 369-371.
32. Siddha Marunthugalin Seimurai, India Maruthuvargal Kooturavu Marunthu Sei Nilayam matrum Pandagasaalai, Chennai-41, 14th Edition, 2003, 175, 177, 224, 284.
33. Agathiyar Vaithiya Kaviyam-1500, 1st Edition, 1994, Tamil PalKalai Kazhagam, Thanjavur-613005, 790.
34. Database On Medicinal Plants Used in Ayurveda, Central Council For Research in Ayurveda and Siddha Jawahar Lal Nehru Bharatiya Chikista Avum Homeopathy Anusandhan Bhawan, New Delhi. Volume I - 73-75, Volume III – 158 & 160, 282 & 284, 472, 474 & 475, Volume IV – 122 & 124, 269, 272 & 273, Volume V – 187, 189 & 190, 315, 317 & 319, Volume VII – 135 to 138, 169 to 171.
35. http://www.phcogrev.com/article, Role of herbs in the management of Asthma, Review Article, 2009, Volume 3, issue 6.
36. Journal of Complementary and Integrative Medicine. Volume 7, Issue 1, Pages –, ISSN (Online) 1553-3840, DOI: 10.2202/1553-3840.1401, September 2010
37. Indian journal of Medical Research
38. Journal of Scientific and Industrial Research, Volume 69, Oct. 2010.
39. Indian journal of Pharmacology, 1994
40. International Journal of Herbal Drug Research, Vol I, Issue 1, 15-18, 2011

41. International journal of pharmacology, 2012, volume 8, issue 3
42. Screening of Indian plants for biological activity.
43. Indian herbal remedies, Rational Western Therapy, Ayurvedic and Traditional Usage, Botany, edited by C.P.Khare, 2004
44. http://en.wikipedia.org/org/wiki/halite.
45. Dr.A.K.Nadkarni, Indian Materia Medica, Volume II, Ramdas Bhatkal for Popular Prakashan Pvt. Ltd, Mumbai.2005, 108.
46. Siddha Materia Medica (Mineral and Animal Kingdom), Department of Indian Medicine and Homeopathy, Chennai-106, 1st Edition, 2008, Dr.Anaivari Ananthan,Ph.D., Dr.M.Thulasimani,M.D.(Pharmaco). 303 & 304.
47. http://www.medscape.com
48. K Sembulingam Prema Sembulingam, Essentials of Medical Physiology, 5th Edition, 2007, JAYPEE Brothers Medical publishers (P) Ltd,78.
49. http://www.botanical-online.com/alkaloids
50. www.medscape.com
51. www.medind.nic.in, Review article, Nutririon and Asthma, 2007, K.B.Gupta, Manish verma.

PHARMACOLOGICAL REFERENCES-DRUG I

1. Bowman WC, Rand MJ (1980). Textbook of Pharmacology. Blackwell Scientific publication, New York pp. 21-31.
2. Clemens MR, Remmer H, Waller HD (1984). Phenylhydrazine-induced lipid peroxidation of red blood cells: in vitro and in vivo monitoring by the production of volatile hydrocarbons. Biochem. Pharmacol. 33: 1715-1718.
3. Dacie JV, Lewis SM (1994). Practical Haematology 8th ed. ELBS, Churchill, Livingstone. pp. 49-59.
4. Diallo A, Gbeassor M, Vovor A, Eklu-Gadegbeku K, Aklikokon K, Agbonon A, Abena AA, de souza C, Akpagana K (2008). Effect of *Tectona grandis* on phenylhydrazine-induced anaemia in rats. Fitoterapia 79(5): 332-336.
5. Dina OA, Adedapo AA, Oyinloye OP, Saba AB (2000). Effect of *T.occidentalis* extract on experimentally induced anaemia in domestic rabbits. Afr. J. Biomed. Res. 3: 181-183.
6. Duff S (2008). Types of Anaemia. www,innvista.com.

7. Erah OP, Asonye CC, Okhamafe OA (2003). Response of Trypanosoma Brucei brucei induced anaemia to a commercial herbal preparation. Afr. J. Biotechnol. 2 (9): 307-311.
8. Muriel C, Jean-Yves LH (1998). Prevalence of and Risk Factors of Anaemia in young children in Southern Cameroon. Am. J. Trop. Med. Hyg. 58 (5): 606-611.
9. Nelson C, Erikson K, Pinero DJ, Beard JL (1997). In Vivo dopamine metabolism is altered in iron deficient anaemic rats. J. Nutr. 127(12): 2282-2288.
10. Oma NU (1991). Iron Deficiency anaemia. Clin. Pharm. Herbal Med. 7 (116): 12-15.
11. Orna NU (1991). Iron deficiency anaemia. Clin. Pharm. Herb. Med. 7: 12-16.
12. Ramzi SC, Vinay K, Stanley LR (1994). Pathologic Basis of Disease, 5th edn. Pub. W.B. Saounders Company. pp. 586-590.

PHARMACOLOGICAL REFERENCES-DRUG 2

1. Abe, T. Yoshida, K. Omata, I. Segawa, Y.Mastuda, K.and Nagai, H. Effects of ZCR 2060 on allergic airway inflammation and cell activation in guinea pigs, J. Pharm. Pharmacol., 46,876 (1994).
2. Abraham, E. Deodhar, M.M. Natu, M.V.A comparative study of the bronchodilator activity of the calcium antagonists on histamine induced bronchospasm in guinea pigs, Indian J. Pharmacol., 24,231-232, (1992).
3. Anilkumar D, Ramu P. (2002). Effect of Methanolic extract of *Benincasa hispida* against histamine & acetycholine induced bronchoplasm in Guinea pigs. Indian J. Pharmacology, 34, pp. 365-366.
4. Anilkumar, D. and Ramu, P. Effect of methanolic extract of *Benincasa hispida* against histamine and acetylcholine induced bronchospasm in guinea pigs, Indian J. Pharmacol,. 34,365-366, (2002).
5. Babe, K.S. and Serafin, W.E. "Goodman and Gilman's The Pharmacological basis of Therapeutics". In; Histamine, Bradykinin and their antagonists, Hardman, J.G. Limdird, L.E. Molinoff, P.B. Ruddon, R.N. and Gilman, A.G. (eds), 9th Edn, Mc GrawHill, New York, 587-588 (1996).
6. Bhujbal SS, Kumar D, Deoda RS, Deore TK, Patil MJ. (2009). Antiasthmatic activity of roots of *Hemidesmus indicus* R. Br. Pharmacologyonline, 1, pp. 209-216.
7. Ghosh, M.N. "Fundamentals of Experimental Pharmacology", Singha.J. (Ed), Calcutta Scientific Book Agency,. Calcutta, 177-82, (1984).

8. Gokhale, A.B. and Saraf, M.N. Bronchoprotective effect of ethanolic extract of *Tephrosia purpurea invivo,* Indian Drugs., 37(7), 346-347, (2000).
9. Govindan SS, Viswanathan S. (1999). A Pilot Study on Clinical Efficacy of *Solanum xanthocarpum* and *Solanum trilobatum* in Bronchial Asthma. Journal of Ethnopharmacology, 66, pp. 205-210.
10. Kumar D, Bhujbal SS, Deoda RS, Mudgade SC. Bronchodilator activity of aqueous extract of stem bark of *Ailanthus excelsa* Roxb. Phcog Res., 2, 102-106.
11. Nair, A.M. and Saraf, M.N. Inhibition of antigen and compound 48/80 induced contractions of guinea pig trachea by the ethanolic extract of leaves of *Virtex negundo* Linn, Indian J. Pharmacol., 27, 230-233, (1995).
12. Saraf MN, Patwardhan BK. (1988). Pharmacological studies on *Sarcostemma brevistigma* Whight part II. Bronchodilator activity. Indian Drugs, 26, pp. 54-57.
13. Singh S. Agrawal S. (1990). Bronchorelaxant activity of *Belamcanda chinensis* (Adans).; Ind. J. Pharmacol., 22, pp. 107-109.

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The Tamil Nadu Dr. M.G.R. Medical University

69, Anna Salai, Guindy, Chennai-600 032

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DR. MAYILVAHANAN NATARAJAN
M.S.Orth. M.Ch.Orth. (L'pool) Ph.D. (Orth. Onco.) F.R.C.S. (Eng) D.Sc.
7th VICE CHANCELLOR

Dr. R. SRILAKSHMI, DCH, Ph.D.
REGISTRAR

Dr. N. KABILAN, M.D. (Siddha)
READER, DEPT. OF SIDDHA

NATIONAL INSTITUTE OF SIDDHA

(An Autonomous Body under Department of AYUSH)
Ministry Of Health & Family Welfare, Government of India

Tambaram Sanatorium, Chennai - 600 047
Tel : 044-22411611 Fax : 044-22381314
E-mail : nischennaisiddha@yahoo.co.in
Website : www.nischennai.org

Name: Dr. T. SATHYA MAHESWARI, REG. NO : 32101706
Title: pre-clinical and clinical study on "SINGATHI CHOORANAM" for "BRONCHODILATOR ACTIVITY" in the management of Eraippu (Bronchial asthma).
No. NIS/IEC/2011/3/146 – 24/12/2011

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has been approved by the IAEC.

Prof. Dr. K. Manickavasakam	Dr. B. Jayachandran Dave
Name of Chairman/Member Secretary IAEC:	**Name of CPCSEA nominee:**
Signature with date	
Chairman/Member Secretary of IAEC:	**CPCSEA nominee:**

(Kindly make sure that minutes of the meeting duly signed by all the participants are maintained by Office)

NATIONAL INSTITUTE OF SIDDHA

(An Autonomous Body under Department of AYUSH)
Ministry Of Health & Family Welfare, Government of India

Tambaram Sanatorium, Chennai - 600 047
Tel : 044-22411611 Fax : 044-22381314
E-mail : nischennaisiddha@yahoo.co.in
Website : www.nischennai.org

Name: Dr. T. SATHYA MAHESWARI, REG NO: 32101706
Title: pre-clinical and clinical study on "VALENDRAPHOLA CHOORANAM" for "HAEMATINIC ACTIVITY" in the management of pandu (Anaemia).
No. NIS/IEC/2011/3/14a - 24/12/2011

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Prof. Dr. K. Manickavasakam	Dr. B. Jayachandran Dare
Name of Chairman/Member Secretary IAEC:	**Name of CPCSEA nominee:**
Signature with date	
K. Manickavasakam	B. Dare
Chairman/Member Secretary of IAEC:	**CPCSEA nominee:**

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Name of Member Secretary IAEC:

Dr. J. Anbu

Name of CPCSEA nominee:

Dr. K. Sadhasivan Pillai

Signature with date

08/08/12

Member Secretary of IAEC

Dr. J.ANBU, M.Pharm., Ph.D., D.M.L.T.,MBA.
Professor & Head
Department of Pharmacology & Toxicology
School of Pharmaceutical Sciences
Vels University
Pallavaram, Chennai-600 117.

CERTIFICATE

This is to certify that the project title: "Preclinical study on "Valendraphola Chooranam" for haematinic activity in the management of Pandu (Anaemia)" has been approved by the IAEC with the reference number. XIII/VELS/PCOL/38/2000/CPCSEA/IAEC/08.08.12

Name of Member Secretary IAEC:

Dr. J. Anbu

Name of CPCSEA nominee:

Dr. K. Sadhasivan Pillai

Signature with date

08/08/12

Member Secretary of IAEC

Dr. J.ANBU, M.Pharm., Ph.D., D.M.L.T.,MBA.
Professor & Head
Department of Pharmacology & Toxicology
School of Pharmaceutical Sciences
Vels University
Pallavaram, Chennai-600 117.